

Proceedings of the Twelfth
Annual Aquatic Toxicity
workshop:
November 5-8, 1985
Thunder Bay, Ontario

Comptes rendus des communications
du douzième atelier annuel sur
la toxicité aquatique :
du 5 au 8 novembre 1985
Thunder Bay (Ontario)

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G. W. Ozburn

July 1986

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Editor/Editeur

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ORGANIZING COMMITTEE

CHAIRMAN: George W. Ozburn, Department of Biology, Lakehead University, Thunder Bay, Ontario, P7B 5E1.

CO-CHAIRMAN: J. Howard McCormick, U.S. EPA, Envir. Res. Lab - Duluth, 6201 Congdon Blvd., Duluth, Minnesota, 55804.

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The organizers wish to thank the following for fulfilling the duties of sessional chairmen: W. Vrooman, W.M.J. Strachan, Gary Westlake, Peter Wells, N. Bermingham, A. Smith, and W.T. Momot.

In addition, thanks are due to the Northwest Region office of the Ministry of the Environment, Aquatic Toxicity Research Group, Lakehead University, the Department of Fisheries and Oceans and the Environmental Research Laboratory, Duluth, Minnesota for helping to make this Workshop a success. The staff of the Valhalla Inn, Thunder Bay, are to be congratulated for the magnificent meals and fine co-operation in setting up for this Workshop. Finally, without participants, there would be no workshop; therefore, the many authors and participants deserve a special thank you for their contribution to success.

A very special thank you is extended to Dr. Donald Mount for taking time from a very busy and demanding schedule to give us an overview of the real Aquatic Toxicity problem after the Banquet.

PREFACE

This report is the Proceedings of the Twelfth Annual Aquatic Toxicity Workshop, held in Thunder Bay, Ontario, from November 5-8, 1985.

The Aquatic Toxicity Workshop is one of a continuing series of annual workshops in Canada on aquatic and environmental toxicology, covering topics from the principles of aquatic toxicology to applications in environmental effects monitoring, setting of toxicity criteria in regulations and guidelines, and the development of water quality objectives. The Workshop emphasizes an informal exchange of ideas and knowledge on the topic among interested persons from industry, governments, consulting firms and universities. The Workshop provides an annual focus in Canada on the principles and approaches in aquatic toxicology, and the role of aquatic toxicology in the prevention and control of water pollution.

The Workshop is run by an incorporated National Steering Committee, and the proceedings are published annually with the support of the Department of Fisheries and Oceans.

Papers and posters were solicited in topics related to research in aquatic toxicology, with emphasis in the following (not prioritized): Site-Specific Pollution Regulation; Environmental Impact Statement; Applicability of Laboratory Studies for Predicting Field Events; The Role of Modelling in Risk Assessment; and Criteria Development.

EDITOR'S COMMENTS

This volume contains the papers and abstracts of papers presented at the Twelfth Aquatic Toxicity Workshop, together with author and subject indexes and a list of participants.

The submitted papers and abstracts were published after external review. Comments or further discussion on any aspect of the contributions should be directed to the authors.

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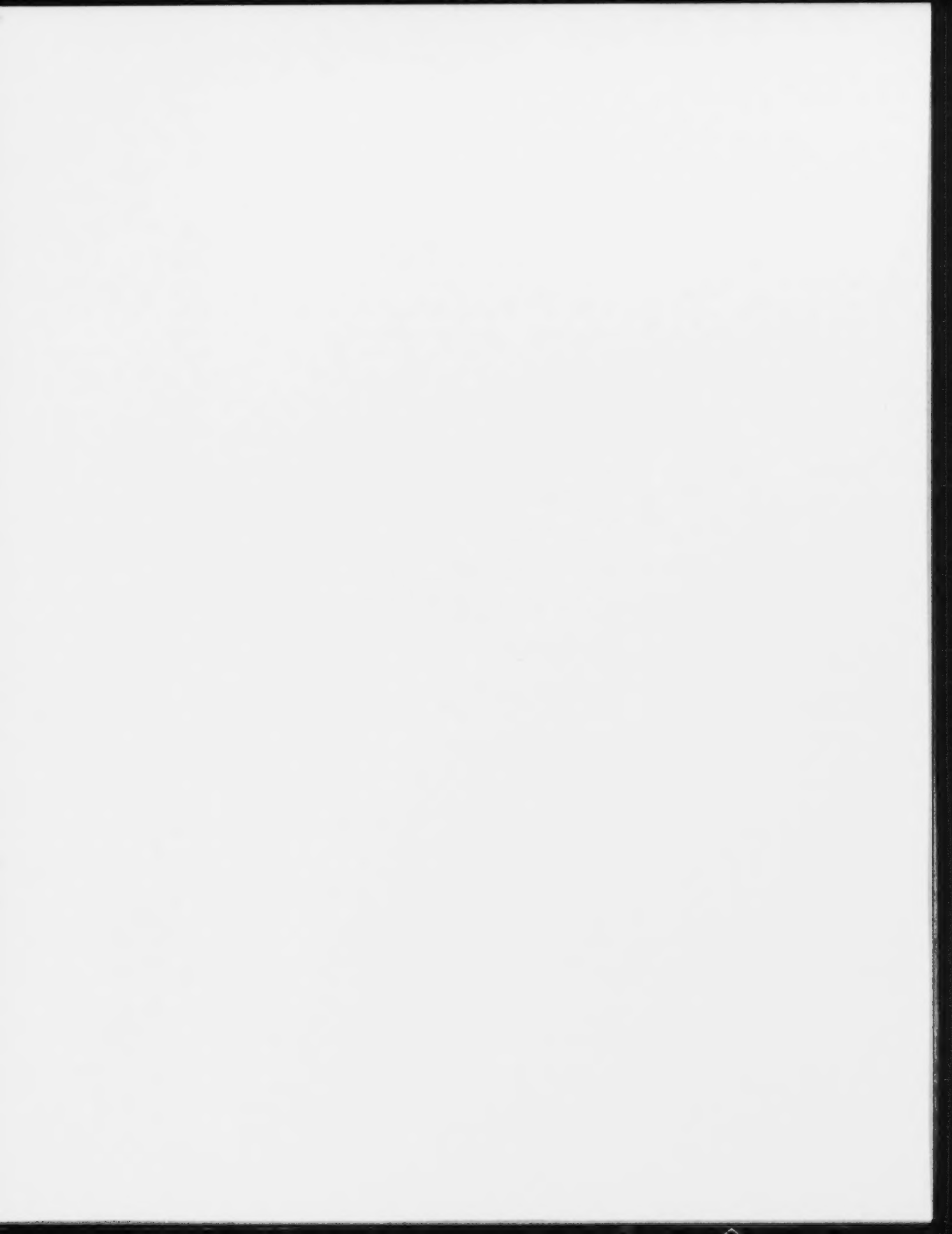
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DULUTH FIELD TRIP REPORT

Twenty participants of the 12th Annual Aquatic Toxicity Workshop were treated to an open house and tour of the U.S. Environmental Protection Agency's aquatic toxicity laboratory, the Environmental Research Laboratory-Duluth, in Duluth, Minnesota. During the morning of Nov. 5, participants were given an overall view of the mission of the laboratory by its director, Dr. Norbert Jaworski. Branch supervisors also gave short synopses of work in their respective fields of pesticides, toxic substances, hazardous wastes, and water quality. Also included was a slide show on the Monticello Ecological Research Station in Monticello, Minnesota.

After a catered lunch, tour guides escorted participants in a 1½ hour walking tour of the various laboratories, allowing time for visitors to ask questions and interact with scientists at work in their laboratories. Tour stops included laboratories studying structure/activity relationships, microcosms, behavioral responses of fish to toxicants, dioxin tissue residue studies, acid rain, pharmacokinetics, pesticide field applications, and the fathead minnow culture unit, which supplies embryos, larvae, and 30 day old fathead minnows for testing throughout the laboratory. Another highlight was the complex effluents field testing trailer, a mobile laboratory in a semi-truck trailer. This mobile laboratory has been used in effluent field studies all over the U.S., and has recently returned from a month of field studies in Sweden. A period of time was open after the tour to allow visitors to return to speak with investigators in fields of particular interest to them. The day's activities were topped off by a fine supper at the Harbor House restaurant in Two Harbors, Minnesota, along the scenic route back to Thunder Bay on the north shore of Lake Superior.



SITE-SPECIFIC POLLUTION REGULATION:
A "ZERO-BASE" PERSPECTIVE

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ABSTRACT

When freed from the constraints of existing toxicant regulations, conventional endpoints of toxicity tests, disciplinary biases, and other (often artificial and arbitrary) ties to history, one can take a fresh look at what we mean when we seek to regulate a specific effluent at a specific location. As in zero-base budgeting, one asks fundamental questions about what is needed.

Discharge of wastewater to aquatic habitats is visualized as a dynamic mixing process that depends on the engineering of the outfall, the hydrodynamics of the specific receiving water, losses of pollutants (e.g., to air or sediment), and physical and chemical reactions in the water, all of which yield predictable concentrations of the pollutants at specific locations and times. The acceptability of these estimated concentrations in relation to maintaining aquatic life in the water body will provide feedback to the selection of allowable concentrations in the wastewater and the design and siting of its discharge. Site-specific factors influencing acceptability include the local species assemblage, the duration of toxicant exposure, and environmental conditions that modify

toxicity (e.g., temperature, oxygen, water hardness). The realities of local sites must be considered, including exposures to fluctuating toxicant concentrations and mixtures of chemicals. How to use generic and laboratory-derived data in site-specific analyses is a major contemporary challenge. Some conventional approaches can still be used, and others can be modified fruitfully.

The principal "emergent property" of site-specific pollutant regulation is the requirement for multidisciplinary cooperation among facility designers, aquatic chemists, hydraulic engineers, limnologists/aquatic ecologists, toxicologists, and modelers in developing environmentally benign wastewater disposal. The regulation of thermal effluents is used as an example of successful application of site-specific assessment and regulation.

Research sponsored jointly by the U. S. Environmental Protection Agency under Interagency Agreement 40-1629-85 and the Office of Health and Environmental Research, U. S. Department of Energy, under contract No. DE-AC05-84OR21400 with Martin Marietta Energy Systems, Inc.

REGLEMENT SUR LA POLLUTION SPECIFIQUE AU SITE UNE PERSPECTIVE A BASE ZERO

par

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SOMMAIRE

Lorsque libéré des contraintes des réglementations existantes sur les produits toxiques, des points terminaux classiques des tests de toxicité, des partis-pris disciplinaires, et des autres liens (souvent artificiels et arbitraires) avec l'histoire passée, on peut jeter un coup d'oeil original sur ce que l'on veut dire lorsqu'on essaye de promouvoir une réglementation sur un effluent spécifique en un emplacement spécifique. Comme pour un budget à base zéro, on soulève des questions fondamentales sur ce qui est requis.

La décharge des eaux usées dans des habitats aquatiques est considérée comme un processus de mélange dynamique dépendant de l'ingénierie de l'issue, de l'hydro-dynamique de l'eau de réception spécifique, des pertes de produits polluants (par exemple, à l'air ou par sédimentation), et des réactions chimiques et physiques dans l'eau, qui produisent toutes des concentrations prévisibles de produits polluants en des emplacements et en des instants spécifiques. L'acceptabilité de ces concentrations estimées en ce qui concerne le maintien de la vie aquatique dans l'eau de déversement prduit des informations permettant de choisir les concentrations acceptables dans les eaux usées et la conception et le positionnement de leurs décharges. Les facteurs spécifiques au site influençant l'acceptabilité

comprennent l'ensemble local des espèces, la durée de l'exposition aux produits toxiques, et les conditions ambiantes qui modifient la toxicité (par exemple, la température, la teneur en oxygène, la dûreté de l'eau). Les réalités des sites locaux doivent être prises en considération, y compris les expositions à des concentrations variables en produits toxiques et aux mélanges de produits chimiques. L'utilisation de données génériques et dérivées du laboratoire pour des analyses spécifiques au site constitue un problème contemporain majeur. On peut encore utiliser certaines approches classiques, et d'autres peuvent être modifiées avec succès.

La principale "propriété émergente" des réglementations sur la pollution spécifique au site réside dans la nécessité d'une coopération multi-disciplinaire entre les concepteurs de l'installation, les chimistes aquatiques, les ingénieurs hydrauliciens, les écologistes aquatiques/limnologistes, les toxicologistes, et les modélistes pour arriver au développement d'une décharge d'eaux usées sans effets sur l'environnement. La réglementation des effluents thermiques est utilisée comme constituant un exemple d'application avec succès d'une évaluation et d'une réglementation spécifiques au site.

Recherche supportée conjointement par l'Agence Américaine pour la Protection de l'Environnement (USEPA) dans le cadre de l'Accord Inter-Agences N°40-1629-85, et par le Bureau de Recherche sur la Santé et l'Environnement (OHER) du Ministère Américain de l'Energie (USDoe) dans le cadre du contrat N° DE-AC05-84OR21400 avec Martin Marietta Energy Systems Inc.

THE INFLUENCE OF DIET ON THE TOLERANCE OF WATERBORNE SODIUM
PENTACHLOROPHENATE BY RAINBOW TROUT (*SALMO GAIIRDNERI* R.).

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University of Waterloo, Waterloo, Ontario

HICKIE, B. E. and D. G. DIXON. 1985. The influence of diet on the tolerance of waterborne sodium pentachlorophenat by rainbow trout (*Salmo gairdneri* R.). Can. Tech. Rep. Fish. Aquat. Sci.

The effects of practical diets, varying in the carbohydrate:lipid ratios, on the acute and chronic toxicity of waterborne sodium pentachlorophenat (NaPCP) to rainbow trout were studied. Fish were raised on one of three isocaloric, isonitrogenous diets and exposed to 0 or 50 ug/L NaPCP. Fish raised on the high-carbohydrate (HC; 20% cerelose, 5% lipid) showed reduced weight gain, elevated liver glycogen content, enlarged livers, a larger liver protein pool and smaller lipid reserves relative to fish raised on the low-carbohydrate diet (LC; 0% cerelose, 14% lipid). The incipient lethal level (ILL) of NaPCP was affected by both diet and pre-exposure to NaPCP. Fish raised on the HC diets tended to be more tolerant of NaPCP than fish raised on the other diets. Pre-exposure to NaPCP reduced the NaPCP tolerance of fish raised on the LC and intermediate (INT; 10% cerelose, 9% lipid) diets. The ILL values were significantly correlated to liver somatic index, liver glycogen, liver protein content and whole fish lipid content. Over a 12-wk exposure to NaPCP, the weight gain of NaPCP LC fish was 25% less than the controls, while weight gain of the NaPCP INT and NaPCP HC fish were only nominally less than controls. Other effects of chronic NaPCP exposure common to all diet groups included decreased liver glycogen reserves and an increase in the liver protein pool size. An interaction between diet and NaPCP exposure affected the lipid content of fish. The influence of these diets on the toxicity of NaPCP to trout may be explained by the effects of lipid pool size on the uptake kinetics of NaPCP.

Key Words: sodium pentachlorophenat, nutrition, tolerance, rainbow trout, *Salmo gairdneri*.

HICKIE, B. E. and D. G. DIXON. 1985. L'influence de regime sur la tolerance de pentachlorophenat de sodium hydrique pour la truite arc-en-ciel (*Salmo gairdneri* R.). Can. Tech. Rep. Fish. Aquat. Sci.

Les effets de régime, dont le rapport entre les hydrates de carbone et graisse varait, sur la toxicité chronique et aigue du pentachlorophenat de sodium hydrique (NaPCP) pour la truite arc-en-ciel (*Salmo gairdneri*) a été étudié. Les truites ont été élevées sur un des trois régimes isocaloriques et isoazotes et exposées au NaPCP (0 ou 50 ug/L). Par rapport aux truites maintenues à un régime pauvre en hydrates de carbone (LC; 0% cerelose, 14% graisse), les truites élevées sur un régime riche en hydrates de carbone (HC; 20% cerelose, 5% graisse) ont présenté une réduction du gain de poids, une teneur élevée en glycogène hépatique, des foies dilatés, une augmentation des protéines hépatiques,

ainsi que des réserves de graisse plus petites. Le niveau letale de NaPCP était influencé par le régime et la pre-exposition au NaPCP. Les poissons maintenus au régime HC avaient la tendance d'être plus tolérants au NaPCP que les poissons maintenus aux autres régimes. Pre-exposition au NaPCP réduisait la tolérance au NaPCP chez les poissons élevés sur les régimes LC et intermédiaire (INT; 10% céréales, 9% graisse). Les valeurs de ILL étaient liées de façon significative à la teneur en protéines hépatiques, à la teneur en graisse du poisson entier. Pendant une exposition au NaPCP de 12 semaines, le gain de poids des individus NaPCP LC était 25% moins que le groupe témoin, alors que le gain de poids des individus NaPCP INT et NaPCP HC était à peine moins que le groupe témoin. Exposition chronique au NaPCP a entraîné une baisse nette des réserves de glycogène dans le foie et une augmentation de la teneur en protéines hépatiques chez tous les groupes. Une interaction entre le régime et l'exposition au NaPCP influait la teneur en graisse des poissons. L'influence de ces régimes sur la toxicité de NaPCP chez la truite peut être expliquée par les effets de la teneur en graisse sur les dynamiques de NaPCP.

EXTENDED ABSTRACT

Laboratory tests with a variety of species are used to assess the toxicity of environmental contaminants to aquatic organisms. While there are detailed descriptions of methodology for both acute and chronic studies, little consideration has been given to the composition of diets fed before or during tests. This research was undertaken to determine if diets varying in the ratio of digestible carbohydrate: lipid, but equivalent in protein and metabolizable energy content (Table 1), would affect the acute and chronic toxicity of sodium pentachlorophenate (NaPCP) to rainbow trout (*Salmo gairdneri*).

The research was approached in two stages. The first involved raising juvenile rainbow trout (initial mean wt 2.5g) on each of the three test diets for 15 weeks (6 tanks of 70 fish per diet). During the final 26 days of this period 3 tanks of fish from each diet group were exposed to 50 ug/L NaPCP. After the 15 weeks fish were sampled in order to determine liver somatic index (LSI), liver protein and glycogen content and whole fish chemical composition. The remaining fish were used to determine NaPCP incipient lethal levels (ILL) for each of the 6 diet-treatment combinations.

In the second stage juvenile rainbow trout (initial mean wt 2.5 g) were exposed to 0 or 50 ug/L NaPCP for 12 weeks to investigate the interactive impact of diet formulation and NaPCP exposure on wet weight gain. The experimental protocol was similar to the one above. The biomass in each tank was determined weekly. At the end of the 12 weeks, samples from each group were taken to determine: LSI, liver protein and glycogen content and whole fish chemical composition.

The conditions under which fish were raised in the two stages were similar. Fish were fed 3-4 times per day at feeding rates based on the method of Hilton and Slinger (1981). Water temperature was maintained at $14.1 \pm 0.3^\circ \text{C}$. Toxicant stock solutions were made with >99% pure

pentachlorophenol (Sigma Chem. Co., St. Louis, Mo.). Water quality parameters were as follows: dissolved oxygen 8.3-9.3 mg/L ; total hardness 395 mg/L as CaCO_3 ; pH 7.7; total alkalinity 267 mg/L as CaCO_3 ; NH_3 6.7 ug/L.

Rainbow trout raised on the high-carbohydrate (HC) diet showed elevated LSI and liver glycogen content, as well as reduced liver protein, whole fish lipid and wet weight gain relative to trout raised on the low-carbohydrate (LC) diet (Table 2, Figure 1).

The ILL of NaPCP to rainbow trout was influenced by the combined effects of pre-exposure to NaPCP and diet (Figure 1). There was no significant difference between the ILLs of the 3 dietary control groups, although there was a tendency for the ILLs to increase from the LC to the HC diet group. This tendency was expressed significantly among the NaPCP pre-exposed groups. The NaPCP pre-exposed fish raised on the LC and intermediate diets were more sensitive to subsequent NaPCP exposure than the HC pre-exposed fish, and all 3 dietary control groups. Pre-exposed fish raised on the HC diet did not differ from the dietary control groups in acute response to NaPCP.

Exposure to NaPCP for 12 weeks directly affected liver protein, liver glycogen content and weight gain (Table 2). There was also some interaction between diet and NaPCP exposure affecting the lipid content of fish. The reduced liver glycogen reserves in NaPCP exposed fish is typical of a chronic stress condition and may lead to greater susceptibility to acute stress. The increase in liver protein content associated with exposure to NaPCP is apparent even when the values are corrected for any changes in liver size associated with reduced glycogen content (liver protein pool). This increase in the liver protein pool size appears to be some adaptation to or compensation for the long-term effects of exposure to NaPCP. While an overall reduction in weight gain due to NaPCP exposure was apparent, a means comparison test (SNK) showed that only the fish raised on the LC diet were significantly impaired. The interaction between diet and NaPCP exposure affecting the lipid content of fish may have been a direct negative effect of NaPCP on lipid reserves which was confounded by the effects of NaPCP on the weight gain of fish.

With recent findings that the lipid content of test organisms affects the apparent bioconcentration factor and uptake kinetics of lipophilic compounds (Chiou, 1985) it is likely that the results from this study can be explained by the effects of the lipid pool size on the kinetics of NaPCP.

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Hilton, J.W., and S.J. Slinger. 1981. Nutrition and feeding of rainbow trout. Can. Spec. Publ. Fish. Aquat. Sci. 55:15 p.

Table 1: Percentage composition and chemical analysis of the test diets. Results of analysis are the mean of the two diet batches used, with 3 samples per batch.

Ingredient	DIET		
	LC	INT	HC
Capelin meal	40	40	40
Soybean meal	20	20	20
Wheat middlings	10	10	10
Wheat gluten	5	5	5
Vitamin pre-mix *	2	2	2
Mineral pre-mix *	1	1	1
Cerelose	0	10	20
Alpha-floc	13	7.5	2
Fish oil	9	4.5	0
Analysis			
Crude protein	42.3	42.1	40.6
Lipid	13.8	9.1	4.7
Glucose	0.6	9.3	18.2
Ash	9.3	8.6	8.6
Crude fibre	10.1	8.2	4.6
Moisture	3.6	3.7	3.5
Gross energy (kJ/g)	21.2	20.0	19.0

* Vitamin and mineral pre-mix as given by Cho et al. (1976).

Table 2: Parameters for rainbow trout raised on the test diets and exposed to 0 or 50 ug/L NaPCP for 12 weeks. Values are means (S.E.M.), n=10 except for weight gain (n=3) and lipid content (n=6). Probabilities from 2-way ANOVA are given below.

Diet	NaPCP conc. (ug/L)	Liver somatic index	Liver glycogen (mg/g liver)	Liver protein (mg/g liver)	Lipid (mg/g fish)	Wt. gain 12-wk (g)*
LC	0	1.29(0.10)	38.3(3.3)	197.8(7.4)	96.3(1.8)	22.7(1.3) ¹
LC	50	1.26(0.05)	26.7(3.7)	229.6(3.7)	106.0(3.3)	17.0(0.9) ^{2,3}
INT	0	1.55(0.09)	51.0(1.4)	179.1(5.4)	88.9(2.0)	20.7(1.4) ^{1,2}
INT	50	1.56(0.06)	35.9(4.2)	219.5(5.3)	85.3(2.3)	19.4(0.9) ^{1,2,3}
HC	0	1.92(0.11)	57.9(3.8)	174.1(6.7)	65.2(1.5)	17.1(0.5) ^{2,3}
HC	50	1.86(0.08)	40.6(2.8)	221.8(4.4)	59.7(2.5)	16.0(0.2) ³
2-WAY ANOVA p						
Diet		0.0001	0.0001	0.0115	0.0001	0.0057
NaPCP		0.6961	0.0001	0.0001	0.9131	0.0046
Diet x NaPCP		0.9309	0.6890	0.3735	0.0047	0.0608

* Values in column with common superscript are not significantly different ($\alpha = 0.05$).

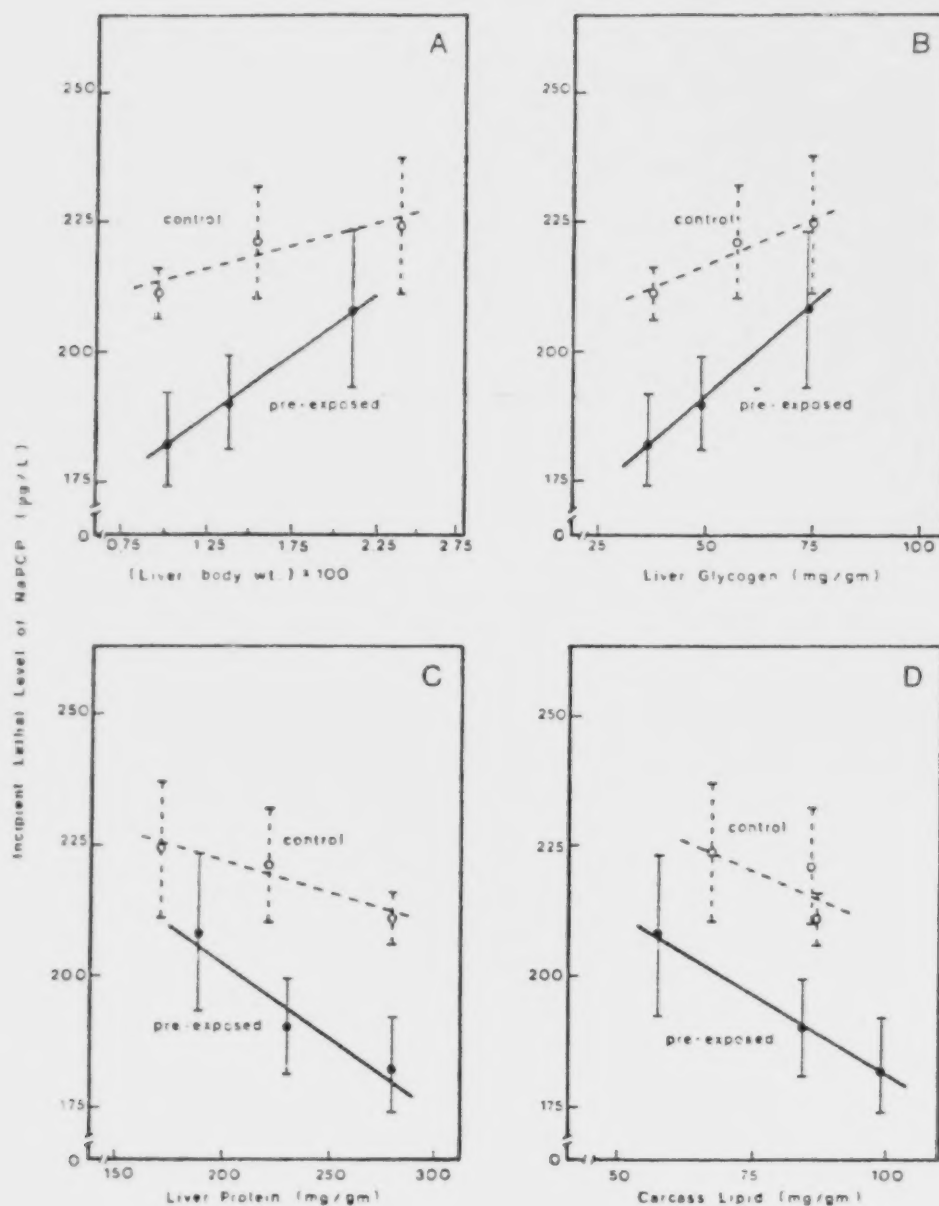


Figure 1. The relationships between four diet dependent parameters and the ILLs for the six treatment groups (control and NaPCP pre-exposed for each of the three diet groups). The error bars represent 95% fiducial limits. A) Liver somatic index vs ILL. B) Liver glycogen per g liver wet wt vs ILL. C) Liver protein per g liver wet wt vs ILL. D) Whole fish lipid content per g wet wt vs ILL.

Bio-availability and toxicity of copper to an indigenous phytoplankton community: the effects of complexation and/or adsorption

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ABSTRACT

In aquatic ecosystems copper toxicity depends on its speciation which in turn, is determined by the presence of organic and inorganic ligands. Apparently, the toxicity of copper to planktonic algae is mostly controlled by the presence of the cupric ion, the toxic form of copper. In this study, we hypothesize that the toxicity of 10, 50 and 100 $\mu\text{g Cu.l}^{-1}$ is lowered by complexation to humic acid and by adsorption to cellulose fibers. This hypothesis was tested in a chemostat with an indigenous phytoplankton community, consisting mainly of diatoms collected from the Saguenay River.

Although the growth (cell division) of all species was inhibited to a certain extent by all copper concentrations used, Asterionella formosa was found to be more sensitive to copper than Tabellaria fenestrata and Melosira islandica, the other dominant species. The rate of primary production and the chlorophyll a content of the community were also negatively affected by copper. The lowering of the rates of photosynthesis as well as of the chlorophyll a content of the cultures accompanied an increase in the copper concentrations used. The presence of humic acid and of cellulose fibers, however, reduced significantly the effects of copper, presumably by lowering availability for the phytoplankton.

We presume that in a river such as the Saguenay, which has a mean copper concentration in the order of 100 $\mu\text{g.l}^{-1}$ and also receives the contaminated effluents of paper mills (cellulose fibers) and of aluminum smelters (metal ions), the complexation of copper to humic acids and/or its adsorption to cellulose fibers are probably significant in reducing the toxicity of copper to the phytoplankton community.

RÉSUMÉ

Dans les écosystèmes aquatiques la toxicité du cuivre dépend de sa spéciation qui est à son tour déterminée par la présence de ligands organiques et inorganiques. En effet, la toxicité du cuivre pour les algues planctoniques est principalement contrôlée par la présence de l'ion cuprique, la forme toxique du cuivre. Dans cette étude, nous avons émis l'hypothèse que la toxicité de 10, 50 et 100 $\mu\text{g Cu}\cdot\text{l}^{-1}$ est réduite par sa complexation à l'acide humique et par son adsorption à des fibres de cellulose. Cette hypothèse a été vérifiée dans un chémostat avec une communauté phytoplanctonique indigène à la rivière Saguenay, constituée principalement de diatomés.

Même si la croissance (division cellulaire) de toutes les espèces a été inhibée jusqu'à un certain point par toutes les concentrations de cuivre utilisées, Asterionella formosa était plus sensible au cuivre que Tabellaria fenestrata et Melosira islandica, les autres espèces dominantes. Le taux de production primaire ainsi que la teneur en chlorophylle a de la communauté ont aussi été influencés négativement par le cuivre. En effet, une réduction des taux de photosynthèse et des concentrations en chlorophylle a des cultures accompagnait une augmentation des concentrations de cuivre utilisées. La présence d'acide humique et de fibres de cellulose a cependant diminué significativement les effets du cuivre sur ces paramètres, présumément en diminuant sa disponibilité pour le phytoplancton.

Nous présumons que dans une rivière comme le Saguenay, où la concentration moyenne de cuivre total est de l'ordre de 100 $\mu\text{g Cu}\cdot\text{l}^{-1}$ et qui reçoit aussi les eaux usées de papeteries (fibres de cellulose) et d'alumine-ries (ions métalliques), la complexation du cuivre aux acides humiques et/ou son adsorption sur des fibres de cellulose réduiraient de façon significative la toxicité du cuivre pour la communauté phytoplanctonique.

THIOCYANATE TOXICITY TO DAPHNIA MAGNA

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ABSTRACT

Thiocyanate is a by-product of the cyanidation process of the gold mining industry and therefore is a contaminant in numerous Canadian lakes. The toxicity of thiocyanate to Daphnia magna was investigated in relation to pH and temperature. Experimental conditions included pH levels of 5, 6, and 7 and temperatures of 8, 12 and 16°. Initial results indicate that at all pH levels, an increase in temperature increases toxicity, whereas an increase of pH decreases toxicity at the temperatures tested. These data show the importance of physical parameters such as pH and temperature on modifying the toxicity of thiocyanate.

Shelley J. Watson and Dr. Edward J. Maly

RESUME

Le thiocyanate est un sous-produit du processus de cyanidation dans l'industrie minière aurifère et par conséquent il est un contaminant de plusieurs lacs du Canada. La toxicité du thiocyanate sur les *Daphnia magna* a été examinée en relation avec le pH et la température. Les conditions expérimentales incluaient des niveaux du pH de 5, 6, 7 et de température de 8, 12 et 16°. Les résultats initiaux indiquaient qu'à tous les niveaux de pH, une élévation de la température accroît la toxicité, alors qu'un accroissement du pH décroît la toxicité aux températures testées. Ces données montrent l'importance des paramètres physiques tel que le pH et la température pour la modification de la toxicité du thiocyanate.

TOXICITY OF INDUSTRIAL EFFLUENTS TOWARDS
RAINBOW TROUT: THE QUEBEC EXPERIENCE.

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Close to 300 effluent samples collected from 160 different industrial sites in the province of Quebec were assessed to determine their acute lethal toxicity to rainbow trout during the 1975 to 1984 period. Chemical analytical support complemented bioassays in an attempt to explain the cause(s) of toxicity. In the six industrial sectors studied, Pulp and Paper ($p < 0.05$) and Food Industry ($p < 0.01$) effluents significantly stood out as the more toxic. Principal component analysis based on a correlation matrix of final effluent data from the Pulp and Paper group pointed to sulfides, phenols, dissolved organic substances, and total solids as toxicity contributors. Results are further discussed in the perspective of prioritizing future actions to improve environmental quality regionally.

LA TOXICITÉ DES EFFLUENTS INDUSTRIELS VIS-A-VIS DE
LA TRUITE ARC-EN-CIEL:
BILAN POUR LE QUÉBEC

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Quelque 300 échantillons d'effluents provenant de 160 différents sites industriels de la province de Québec ont été évalués afin de déterminer leur toxicité létale aiguë envers la truite arc-en-ciel durant la période 1975-84. Des analyses chimiques ont été réalisées en support aux bioanalyses dans le but d'explicitier les causes de la toxicité. Parmi les six secteurs industriels étudiés, ceux des Pâtes et Papiers ($p = 0,05$) et de l'alimentaire ($p = 0,01$) se sont révélés les plus toxiques sur le plan statistique. Pour le secteur des Pâtes et Papiers, une analyse en composantes principales établie à partir d'une matrice de données pour les effluents finaux identifia les sulfures, phénols, matières organiques dissoutes et les solides totaux comme contributeurs importants des effets toxiques observés. L'examen des résultats est intéressant dans la mesure où ceux-ci permettent d'établir un ordre de priorité d'actions correctives à prendre afin d'améliorer la qualité environnementale sur le plan régional.

Development and Validation of Site-Specific Water Quality Criteria for Copper

by

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Running Head: Copper Criteria Validation

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ABSTRACT

Comparative acute toxicity values for copper exposed Ceriodaphnia dubia, Scaphrolebaris sp., and Pimephales promelas were used to calculate mean water effect ratios (e.g., site water LC50 value/reference water LC50) reflective of the difference in the biological availability and/or toxicity of copper between Naugatuck River, Connecticut water and Lake Superior reference water. These ratios were used to modify U.S. Environmental Protection Agency (U.S. EPA) ambient aquatic life criteria for copper to site and station specific criteria using the indicator procedure of the U.S. EPA site-specific guidelines. A mean water effect ratio of 1.0 was obtained using unpolluted upstream water resulting in a site-specific criterion maximum concentration (CMC) and criterion continuous concentration (CCC) of 8.7 and 6.2 $\mu\text{g/l}$ copper, respectively. Mean water effect ratios of 3.9 to 7.0 reflective of reduced biological availability and/or toxicity of copper were determined for four successive downstream stations which contained copper and other industrial and domestic wastes. The resulting station specific CMC(s) and CCC(s) ranged from 32 to 57 and 22 to 39 $\mu\text{g/l}$ copper, respectively. These copper criteria were compared to effluent contributed ambient copper concentrations and ecological survey data from each downstream station to ascertain impact on aquatic life. It was concluded that the national and site-specific criteria derived for copper would be protective of the rivers aquatic life because a relatively healthy aquatic community existed where these criteria were exceeded slightly. Whether or not the station specific criteria were protective could not be determined because these criteria were not exceeded at stations with healthy communities, however, where they were exceeded, impaired aquatic communities were evident.

Generally, C. dubia survival and young production data from receiving water tests and copper addition tests, conducted before the acute toxicity tests were indicative of reduced copper biological availability and/or toxicity in the Naugatuck River at downstream stations.

UPTAKE OF TRACE METALS BY VEGETATION FROM SEWAGE SOURCES

Alfred P. Bernhart* and
Fidenzio G. Salvatori**

Abstract

Over sixty elements have been found to be assimilated by plant tissues. Many of these elements known as trace elements or micronutrients include the trace metals. The micronutrient metals, include but are not limited to iron, manganese, molybdenum, copper, zinc and cobalt all of which constitute an integral part of the plant biomass and are thus required for photosynthesis and various metabolic functions.

Sewage effluent is often rich in trace metals and the opportunity exists for their recovery by plant uptake. Phytotoxicity of trace metals such as copper and zinc is a serious agricultural concern, especially in view of the present popularity in the application of sewage effluent and sludge to farmland. Plants however, have displayed a wide range of tolerance to trace metal accumulation. Vegetable crops are the least tolerant, field crops are intermediate, and grasses and woody plants have a relatively higher tolerance. Harmful effects of trace metals, especially of cadmium, to our food chain is currently of great concern. The concentration of trace elements in plants grown with sewage fertilizers however, is lowest in the fruit or seed and several times higher in the roots, stems and foliage. In addition, phytotoxicity due to trace metals resulting in plant injury or death usually sets in long before plants can accumulate enough metals to be toxic to consumers.

The successful recovery of trace metals by vegetation is largely dependent on the following parameters: the quality of sewage, sewage application modes and rates, soil pH and C.E.C., soil texture, drainage, and plant genotype. Neutral soil pH, well drained fine textured soils, sewage effluent from domestic sources are among some of the ideal conditions for nutrient metal uptake by plants.

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Résumé

Plus de 60 éléments ont été trouvés étant assimilés par le tissu des plantes. Plusieurs de ces éléments connus comme des quantités minuscules ou microéléments inclut la présence de métaux. Les microéléments métalliques, ne sont pas limités au fer, manganèse, molybdenum, cuivre, zinc et cobalt lesquels constituent une partie intégrante de la biomasse végétale et sont ainsi nécessaires à la photosynthèse et aux différentes fonctions métaboliques.

La matière constituante des égouts est souvent riche de ces microéléments et il y a là, une opportunité de les recycler par le cycle de croissance des plantes. La phototoxicologie des microéléments tel le cuivre et le zinc est un sérieux problème en agriculture. Spécialement avec la présente popularité d'application des égouts sur les terres agricoles. Cependant, les plantes ont démontré une grande tolérance à l'accumulation de ces microéléments. Les plantes maraîchères sont moins tolérantes, tandis que les grandes cultures et les plantes ligneuses deviennent progressivement plus tolérantes. Les effets nuisibles de ces microéléments, spécialement le cadmium, dans notre chaîne alimentaire est un grand problème. La concentration de ces microéléments à l'intérieur des espèces cultivées avec les fertilisants des égouts est cependant plus basse dans le fruit ou les graines, et est plusieurs fois plus élevée dans les racines, les tiges et le feuillage. En addition, la phototoxicologie causée par les microéléments métalliques qui crée des dommages ou qui cause la mort aux plantes s'installe habituellement bien longtemps avant que les plantes puissent accumuler assez de métaux pouvant être toxique aux consommateurs.

Le recyclage de microéléments par la végétation est largement déterminé par les paramètres suivants: la quantité d'égouts, les modes et taux d'application des égouts, le pH des sols, C.E.C. la texture des sols, drainage et le génotype des plantes. Les sols avec un pH neutre, les sols bien drainés, les égouts de source domestiques sont parmi quelques-unes des conditions idéales pour utiliser ces métaux nourrissant, servant à la croissance des plantes.

Introduction

The superbly balanced chemistry in nature insures that a multitude of nutrients are continuously being cycled and recycled to and from the abiotic and the biotic environment. This relentless cycling of nutrients is a critical construct of our food chain and bioproductivity. Plants play a major role in the cycling and production of nutrients--indeed they are the chemical factories which transform abiotic material into protoplasm.

Trace metals have an essential role in this nutrient equation, yet they are more often than not cited for their toxic characteristics rather than for their nourishing qualities. Nutrient rich sewage effluent contains significant levels of trace metals; their use as nutrients for plant growth will have the added benefit of sewage treatment and toxicity safeguards. Our attitude towards trace metals as toxins and wastes is part of the problem: wastes are disposed, nutrients are used.

Trace Metals as Micronutrients

Approximately sixty elements have been found to occur in plant tissues (Wilson and Loomis, 1967). Seventeen of these elements have been deemed universally essential for the healthy growth of higher plants (Table 1). Nine elements are used in relatively large quantities and are thus known as macronutrients. The other eight are used in minute amounts hence are known as micronutrients or trace elements. The minor or "non-essential" trace elements found in plant tissue, which are not believed to be critical for survival, may also serve to stimulate plant growth (Brady, 1979). Iron was discovered to be an essential micronutrient in 1847, but the necessity for the others was only established in 1914. As plant nutritional study techniques improve, it is likely that the "non-essential" elements will be recognized as having a more definite role in plant growth.

Eight of the essential nutrients are metals. Magnesium is a macronutrient, while iron, manganese, molybdenum, copper, zinc and cobalt are micronutrients or trace metals. In addition, of the so called "non-essential" elements, there are over 20 trace metals: beryllium, titanium, vanadium, chromium, manganese, iron, cobalt, nickel, copper, zinc, yttrium, zirconium, molybdenum, cadmium, tin, mercury, gold, lead, arsenic, aluminium and selenium. Trace metals are required by plants in minute quantities as compared to the macronutrients. For instance, of the essential elements in alfalfa we find that there are at least 10 million hydrogen atoms for each molybdenum atom. Nevertheless, normal growth would not occur without the presence of molybdenum (Brady, 1974). Molybdenum, as other micronutrients, can be limiting to a whole ecosystem. Goldman (1965) found that the addition of 100 parts-per-million of molybdenum increased the rate of photosynthesis in a mountain lake. In the same lake, however, high cobalt concentrations were inhibiting to phytoplankton. As with all nutrients a surplus, as well as deficiency, can be both equally limiting--albeit the range of concentration of the micronutrients in which plants will grow is smaller than that of the macronutrients (Table 3).

With the exception of iron and at times manganese, the occurrence of trace metals in most soils is rather meagre, hence their availability to plants is low and at times limiting. It is not uncommon then to find soils with a deficiency of trace nutrients. This is in fact the case with agricultural soils whereby the cumulative effect of crop production, enhanced by improved crop varieties and macronutrient fertilizers, has reduced the limited quantities of trace nutrients originally present in the soil (Table 4). This is a case in point where the rich supply of trace metals found in sewage effluent and sludge could be put to good use to restore the balance of essential micronutrients (Table 2).

Biological Role of Trace Metals

Ninety-four to 99.5% of fresh plant tissue is synthesized from nutrients obtained mostly from the air and water: carbon, hydrogen and oxygen. The rest of the essential elements and other "minor" elements comprise only 0.5 to 5 or 6% of the plant tissue. In spite of this apparent imbalance trace elements exert just as much control on plant growth and health as do the macronutrients. This can be explained by Liebig's "law of the minimum", which states that under "steady-state" conditions the growth of plants will be limited by the nutrients most closely approaching the critical minimum limit required for survival. In other words, the level of biomass production cannot be greater than the most limiting of the essential elements.

Although the role of micronutrient metals in plant and microbial growth processes is not entirely understood, many scientists agreed that trace metals are effective through certain enzyme systems. Copper, iron and molybdenum act as "electron carriers" in enzyme systems which bring about oxydation-reduction reactions essential to plant development and reproduction. Copper acts as a catalyst for respiration and for the utilization of iron in chlorophyll synthesis. Magnesium, although not a micronutrient, is also a necessary constituent of chlorophyll, without which green plants could not exist. Zinc and manganese also function in enzyme systems which are required for plant metabolism. Zinc appears to control the formation of growth hormones and the reproduction process of certain plants. Molybdenum is believed to be essential in the process of nitrogen fixation and in the metabolization of nitrates into amino acids and proteins (Brady, 1974). Cobalt is a vital constituent of Vitamin B12 which is believed to be necessary for the formation of a type of hemoglobin in nitrogen-fixing nodule tissue. We can summarize by saying that manganese, iron, zinc and perhaps vanadium are required for photosynthesis. Molybdenum, cobalt and once again iron are required for nitrogen metabolism; others such as manganese, cobalt and copper are required for other metabolic functions (Odum, 1971).

Plant Homeostasis in the Uptake of Trace Metals

The origin of trace and or heavy metals is from the lithosphere. Weathering, plant uptake and decay, plus the strong retention of these metals

by organic and mineral soil colloids increase the concentration of trace metal ions at the soil surface (Table 2). Interestingly enough the range of trace metal concentrations in sewage effluent and sludge are often less than in surface soils, albeit the levels of trace metals can be much higher in primary sludge. The levels in plants however, are considerably less than in surface soils. This can in part be explained by the tight bonding created by the attraction of metal cations to negatively charged hydroxyl and oxygen anions resulting in insoluble and stable hydroxides and oxides. The physiology of plants is also significant here. Plants have a remarkable ability in self-regulating the level of nutrient uptake required--this partially explains why certain plant species can tolerate different levels of available trace metals in soils. As compared to animals, plants had to develop a greater range of tolerance because they are confined by their roots to a limited volume of soil (Bohn et al, 1979).

Experiments carried out to extract the amount of trace metals available to plants from sewage sources using DTPA solution revealed that the amount of extractable metals was very high in comparison to crop uptake (Keeney and Welsh, 1975). In the same experiment it was found that the uptake rate of trace metals by corn grown on two different soil textures (silt loam vs. sandy loam) with a different C.E.C. (22 vs. 13 meq./100g.) was remarkably similar. These two results further verifies the self-regulating nature of plants or the principle of homeostasis: the tendency for biological systems to resist change and to remain in a state of equilibrium.

Phytotoxicity of Trace Metals and Plant Tolerance

Plants in general can tolerate certain ranges of levels of concentration of trace metals in their tissues (Table 3). Cadmium, nickel and lead appear to have the narrowest range of concentration levels. The vegetable crops that are the least tolerant to trace metals: beet, turnip, kale, mustard, lettuce and tomatoes, while beans, cabbage, collards and other vegetables are less sensitive. Field crops such as corn and small grains have an intermediate tolerance while the grasses, such as fescue, lovegrass, Bermuda grass and perennial rye grass are the most tolerant to high metal loadings (Bache et al, 1984). Sopper (1979) further concluded that woody plants, trees and shrubs, are not as sensitive to trace metal toxicity as are agricultural crops.

Generally speaking plants will remove only a small fraction: approximately 5% of the applied heavy metals in waste water. There are however considerable variations. Experiments carried out with sewage sludge indicated that even much more smaller portions were removed by crops (Table 5). Here, however, the results pertain to the removal of the first crop of annual ryegrass, harvested approximately 30 days after seeding. In addition, sewage sludge has a far greater concentration of heavy metals than does sewage effluent, especially at the excessive application rate of 1600 kg.N/ha used in the experiment. Smaller doses (i.e. 400 kg.N/ha) significantly improve removal rates and total biomass (Soon and Bates, 1981).

Effects on the Food Chain

There is a great deal of concern that the uptake of excessive amounts of trace metals by plant roots and subsequent transfer to edible portions of the plant could be hazardous when ingested by primary and secondary consumers. Generally speaking, however, plants have built-in biological barriers which to a large extent prevent the translocation and accumulation of toxic concentrations of trace metals to their edible portions. Experiments have shown that the greater portion of trace metals accumulate in the roots, stems and foliage of plants and less so in the seed and fruit. Work conducted in this area by Keeney and Walsh (1972), (Bernhart, 1985) and Soon and Bates (1981) all confirm this "safeguard" phenomenon of plants. Table 6 dramatizes the relationship between the concentration of trace metals in corn stover and grain. In addition phytotoxicity especially of nickel and copper ensures that plants die or fail to grow long before they can accumulate excess metals toxic to mammalian consumers (Table 3). There are however exceptions, the locoweed plant (*Astragalus* Sp.) absorbs selenium in concentrations that are apparently toxic to browsing animals with no apparent effect on its growth vigour and health (Wilson and Loomis, 1967). Maintaining a cadmium/zinc ratio of 1:100 in sewage effluent or sludge will lead to phytotoxic conditions due to zinc before cadmium zootoxicity threshold are reached in plant tissues (Bache and MacAskill, 1984). Table 7 gives a broad cross-section of the threshold concentration of trace metals for plants, fish and for irrigation and drinking water supplies.

In the same experiment conducted by Keeney and Walsh total metal concentration in the plant tissue increased with increasing rates of sludge applied. There was, however, no significant increase in the corn grain. Soon and Bates (1981) made similar conclusions (Table 8). Keeney and Walsh also demonstrated that the metal concentration even in crops receiving high sludge treatments (60 metric tonnes/ha) were generally within the range found in plant tissues of corn. Sopper and Kerr (1980) attributed this to biological dilution due to enhanced biomass production resulting from waste water irrigation. For example, the total biomass produced by the effluent irrigated reed canarygrass averaged 11.34 metric tonnes/ha as compared to 3.16 MT/ha for the same non-irrigated crop.

In a sixteen year experiment conducted at Penn State University (Sopper, 1979) employing spray irrigation of secondary sewage effluent revealed that the concentration of trace metals in the foliage of herbaceous species remained the same or decreased over the duration of the experiment--only copper showed a slight increase. Also no significant change was detected in the concentration of copper, chromium, lead, cobalt and cadmium within the overall soil profile following a fourteen year period of experimentation (Table 9). Significant increase in zinc and nickel were however recorded at all soil depths up to 30 cm. Nevertheless these increases were still within the range found in Pennsylvania soils.

Factors Controlling Plant Uptake of Trace Metals

The task of employing a biological approach to sewage treatment and uptake of trace metals is a multifaceted one. Many factors and variables are involved including the quality of the sewage effluent or sludge, sewage fertilizer application rates and techniques, soil acidity and cation exchange, soil texture and drainage capacity, and plant genotype. In regards to sewage quality "domestic" waste-water effluent is preferred opposed to sewage sludge--the concentration of trace metals of the latter is much higher (Table 2). To assess the quality and appropriateness of sewage sludge for land application the criteria developed by the Ontario Ministries of Environment and Agriculture and Food can be used (Table 10).

In addition to Table 10 the C.E.C. of the soil also plays a major role in determining the application rate of sludge. For example, rates can be either increased or decreased by a factor of ten with a change of approximately 10 meq./100g. of exchangeable ions (Bache and MacAskill, 1984). The various metal uptake of different plants would also augment the recommended application rates in Table 10. Spray irrigation and overland flow application techniques of sewage effluent and sludge appear to be the two which optimize the plant-soil assimilative capacity (Bache and MacAskill, 1984). Application rates are also of concern since not only can they increase the loading of trace metals but can also depress the soil pH values (Iskandor, 1975). Other modes of treatment including artificial reed marshes and evapotranspiration beds (Bernhart, 1985) may also prove significant in the recovery of trace metals by plants.

The pH of both sludges and soils should ideally be maintained between 7.5-7.0 (Sopper, 1979). Lower pH values will tend to release more metal cations therefore risking the prospect of phytotoxicity, food chain contamination and surface and ground water pollution. Poorly drained or flooded soils also tend to favour soluble cations as compared to well drained soils. Higher pH values are more apt to change cations to insoluble forms, hence making them unavailable to plants. Fine mineral soils and organic soils tend to be highly adsorptive of metal ions, hence reducing the risk of water pollution from surface runoff--the corollary applies to coarse soils.

Conclusions and Recommendations

Davis and Carlton-Smith (1980) examined 39 agricultural crops to assess their comparative efficiencies of assimilating trace metals from soils. In general, however, researchers experimenting with sewage fertilizer have employed only a handful of agricultural crops, and with the exception of Sopper very little work has been done with woody plants. Further research involving the uptake of trace metals by woody plants seems to be a timely task, in particular because trees appear to be less sensitive to trace metal toxicity than are agricultural crops. For this reason the authors have engaged the Ontario Ministry of Natural Resources in establishing two on-site sewage effluent recovery units to monitor the uptake of essential plant nutrients including trace metals by woody plants--full report to be published in 1989.

Although plants absorb rather low quantities of trace elements, it remains valid to suggest that vegetation can provide a useful means of buffering the environment from available forms of trace elements. In addition the employment of trace metal nutrients from sewage sources for plant propagation in agriculture and forestry could have a significant economic benefit.

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Table 1. Essential Plant Nutrients and Other Trace Elements Found in Plant Tissue

Macronutrients	Micronutrients	Other Trace Elements
Carbon	Iron	Beryllium
Hydrogen	Manganese	Aluminum
Oxygen	Boron	Titanium
Nitrogen	Molybdenum	Vanadium
Phosphorus	Copper	Chromium
Potassium	Zinc	Nickel
Calcium	Chlorine	Cadmium
Magnesium	Cobalt	Tin
Sulfur		Mercury
		Lead
		Arsenic
		Selenium

Table 2. Typical Averages and Ranges of Metal Concentrations

Metal Elements	Lithosphere Occurrence (ppm)	Range in Soils (ppm)	Sewage Effluent (ug/l)	Secondary Sludge (ppm)	Plant Biomass (ppm)
Beryllium (Be)	6	3-40	?	?	?
Magnesium (Mg)	?	200-1500	700-1500	500-1000	2000-7000
Aluminum (Al)	?	?	0-15	110-5300	.000001
Titanium (Ti)	4400	1000-10,000	?	?	?
Vanadium (V)	150	20-500	0-01	?	0-0.005
Chromium (Cr)	200	5-1000	20-120	3.3-450	1-5
Manganese (Mn)	1000	200-2000	2-10	4.6-36	0.1-0.3
Iron (Fe)	50,000	50,000-300,000	300-600	540-360	20,000-300,000
Cobalt (Co)	40	1-70	40-80	0.0-2.2	1-40
Nickel (Ni)	100	10-1000	50-100	1.0-209	0.5-6
Copper (Cu)	70	2-100	50-100	18-105	4-20
Zinc (Zn)	80	10-300	30-300	40-680	15-18
Arsenic (As)	?	?	0.01-1.0	?	.0006
Selenium (Se)	?	?	0.01-1.0	.01-.11	.000002
Yttrium (Y)	30	3-80	?	?	?
Zirconium (Zr)	220	60-2000	?	?	?
Molybdenum (Mo)	2	0.2-5	100-500	0.0-3.0	0.5-4
Cadmium (Cd)	?	0.01-7	3-10	0.20-4.10	0.04-0.8
Tin (Sn)	40	?-15	0-0.05	?	0.001
Lanthanides	?	10-500	?	?	?
Mercury (Hg)	?	0.02-0.2	3-5	0.13-0.59	0.02-0.1
Lead (Pb)	11	2-200	100-200	5.6-107	4-6

? no data available

* compiled from a number of sources including Bohn et al (1979), Brady (1974), Bernhart (1985) and Soon et al (1981).

Table 3. Critical Concentration of Trace Metals in Plant Tissue*

Threshold	Cd		Cu		Ni		Pb		Zn		Mn		Mo	
	**	***	**	***	**	***	**	***	**	***	**	***	**	***
Phytotoxicity	8	0.1-1	50	30	11	50	35	5	200	500	0.3	100	4	?
Deficiency	?	?	4	10	?	?	?	?	20	50	0.1	?	0.5	?

* after Boche and MacAskill (1984)

? no data available

** phytotoxic thresholds

*** zootoxic thresholds

Table 4. Areas of Trace Metal Deficiencies, Range in Recommended Rates of Application in the Deficient Areas, and Some Crops Having a High Requirement for Trace Metal Nutrients.*

Metal Micronutrient	Area Deficient in U.S.A. (millions of hectares)	Application Rates (kg. of metal/ha)	Crops Having a High Requirement
Iron	1.54	0.5-10	blueberries, cranberries, rhododendron, peaches, grapes, nut trees
Manganese	5.26	5.30	dates, beans, soybeans, onions, potatoes, citrus
Zinc	2.63	0.5-20	citrus and fruit trees, soybeans, corn, beans
Copper	0.24	1.20	citrus and fruit trees, onions, small grains
Molybdenum	0.61	0.05-1	alfalfa, sweet clover, cauliflower, broccoli, celery

* after Brady, (1974).

Table 5. Ranges of Metal Additions and First Crop (ryegrass) Removal by the Application of 1600 Kg N/ha of Sewage Sludge from 9 Different Sources in S. Ontario.*

Trace Metal	Added to Soil (Kg/ha)	Removed by Crop (g/ha)	% Removed of Total Applied
Cd	0.2-7.1	1.6-7.5	.8-.9
Cr	2.1-310	3.2-8.0	.1-.003
Cu	11-201	31-83	0.3-.04
Ni	0.4-473	7.4-183	1.8-.03
Pb	1.5-116	9.2-17	0.2-.01
Zn	11-390	66-916	0.6-.2

* after Soon and Bates. (1981).

Table 6. Average Trace Metal Concentrations in Corn Stover and Grain Following Application of Sewage Sludge. *

Trace Metal	Corn Stover	Corn Grain
Cd	0.21	.09
Cu	90.83	26.17
Ni	3.48	.56
Zn	.98	1.26

* after Keeney and Walsh (1975).

Table 7. Threshold Concentration (p.p.m.) of Trace Metals for Irrigation Water (i.w.), drinking water (d.w.), drinking water for livestock (d.w.l.) and for plants and fish toxicity.

Trace Metals	AWRC (i.w.)	EPA (i.w.)	ESB (d.w.l.)	EPA (d.w.l.)	WHO (d.w.)	Toxicity* to plants	Toxicity** to fish
Al	1.0	5.0	5.0	?	?	0.43	1.5
V	?	?	0.10	?	?	?	?
Cr	?	0.1	1.0	1.0	0.05	3.8	120
Mn	2.0	0.2	?	?	0.5	0.05	100
Fe	?	5.0	?	?	1.0	4.3	250
Co	?	?	1.0	?	?	?	?
Ni	0.5	0.2	?	?	?	0.18	30
Cu	0.2	0.2	0.5	0.5	1.5	0.02	0.02
Zn	5.0	2.0	24	25	15	1.3	1.3
Cd	0.005	0.01	0.05	0.05	0.01	2.1	2.1
Hg	?	?	0.01	?	0.001	6.6	9.5
Pb	?	5.0	0.1	0.1	0.1	1.7	1.7

AWRC Australian Water Resources Commission

EPA Environmental Protection Agency (U.S.)

ESB Environmental Studies Board (Eng.)

WHO World Health Organization

* based on experiments with Lolium perenne

** based on experiemtns with rainbow trout (*Salmo gairdneri*)

? no data available

Table 8. Metal Concentration in Four Crop (harvests) of Ryegrass Receiving Sewage Sludge From Selective Sources in S. Ontario.*

	Cadmium		Copper		Nickel		Zinc	
	**	***	**	***	**	***	**	***
First	3.30	3.30	42	42	46.8	46.8	319	319
Second	2.45	3.68	36	40	47.7	44.8	249	206
Third	2.26	2.70	16	28	36.8	34.8	216	242
Fourth	1.87	3.12	16	35	40.6	46.0	121	159
TOTAL Removed (ug/g)	9.88	12.8	110	145	171.9	172.4	905	926
TOTAL Applied (kg/ha)	7.0	34.2	201	596	473	1528	387	2576

* after Soon and Bates (1981)

** sludge applied once only at a rate of 1600 Kg N/ha

*** sludge applied and reapplied at a rate of 1600 Kg/ha before seeding of each crop

Table 9. Extractable Trace Metal Concentrations (ug/g) in the Surface 30 cm of Soil in the Penn State White Spruce--Old Field Forest Ecosystem for 14 Years with Municipal Waste Water.*

Year	Cu	Zn	Cr	Pb	Co	Cd	Ni
Irrigated**							
1963	0.65	3.23	0.09	4.61	1.80	0.04	0.56
1965	0.95	3.78	0.06	4.21	2.75	0.04	0.67
1967	1.43	6.15	0.04	4.45	3.21	0.07	0.89
1971	1.23	6.01	0.07	4.19	3.73	0.05	0.54
1976	1.92	7.48	0.01	3.29	1.87	0.03	0.73
Control							
1963	0.93	2.45	0.07	2.99	1.23	0.05	0.32
1965	0.66	2.63	0.08	3.76	2.12	0.05	0.28
1967	1.16	1.93	0.08	3.66	1.81	0.03	0.30
1971	0.92	3.91	0.10	3.69	1.43	0.06	0.35
1976	2.49	2.85	0.10	3.75	0.70	0.07	0.88

* after Sopper (1979)

** application rate of 5 cm/wk during growing season

Table 10. Metal Criteria for Sewage Sludge Application*

	As	Cd	Co	Cr	Cu	Hg	Mo	Ni	Pb	Se	Zn
Content (a) in Ontario Soils (mg/l)	7	0.8	5	15	25	0.1	2	16	15	0.4	55
Max. (b) Content in Soil (mg/l)	14	1.6	20	120	100	0.5	4	32	60	1.6	220
Max. (b) Addition to Soil (Kg/ha)	14	1.6	30	210	150	0.8	4	32	90	2.4	330
Min. (c) Nitrogen to Metal Ratio in Sludge	100	500	50	6	10	1500	180	40	15	500	4
Max. (d) Sludge Applications	10	6	11	9	11	9	5	9	10	9	10
Max. (e) Nitrogen Metal Ratios	480	4200	220	32	45	8400	1700	210	75	2800	20

* after Ontario Ministries of Environment and Agriculture and Food

(a) mean metal content in uncontaminated soils

(b) maximum recommended

(c) minimum ammonium plus nitrate nitrogen to metals ratios

(d) number of sludge applications to achieve the recommended metal content in soil, based on 135 kg. of ammonium plus nitrate nitrogen per application and sewage sludge having minimum ratios

(e) maximum ammonium plus nitrate nitrogen to metals ratios to achieve the maximum recommended metal content in soil

Influence of Chloride on the Availability of Mercury For Bioaccumulation

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MERKOWSKY, A.T., U.T. Hammer, and P.M. Huang¹. Influence of chloride on the availability of mercury for bioaccumulation.

Total mercury concentrations in water declined over a 10-14 day period, from high initial levels (56 ppb) in artificially treated systems to low levels (0.2-0.6ppb) similar to those found in the controls (no mercury added). This occurred in systems with either low (12-18 ppm) or high (150-180ppm) concentrations of chloride in water over lake sediments and in systems with low chloride levels in water over sand substrate. At high chloride levels in water over sand the mercury concentration in water declined only to 2.0 ppb. Uptake of mercury by sediments was related to the percentage of organic content and particle size of the sediments and the chloride levels of the water.

Amphipods (Hyalella azteca) and the rainbow trout (Salmo gairdneri) exposed to systems with added mercury all exhibited elevated mercury levels.

Key Words: Mercury, Hyalella azteca, Salmo gairdneri, chloride, sediments

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Durant une espace de 10-14 jours, les concentrations totales en mercure dans l'eau ont baissé d'une haute teneur initialement (56 ppb) dans les systèmes traités artificiellement, à une faible teneur (0.2-0.6 ppb) semblable à celles trouvées dans les groupes témoins (pas de mercure ajouté). Cela a eu lieu dans les systèmes ayant des concentrations en chlorure, soit faibles (12-18 ppm) ou hautes (150-180 ppm), dans l'eau sur les sédiments lacustres, ainsi que dans les systèmes de faible teneur en chlorure dans l'eau sur un substrat sablonneux. À une haute teneur en chlorure dans l'eau sur le sable, la concentration de mercure dans l'eau baissait à seulement 2.0 ppb. L'assimilation du mercure par les sédiments était relatif à le pourcentage du contenu organique et la taille des particules sédimentaires, ainsi que la teneur en chlorure de l'eau.

Les amphipods (Hyalella azteca) et les truites arc-en-ciel (Salmo gairdneri) exhibaient tous une teneur en mercure élevée quand exposés à un système où on a ajouté du mercure.

INTRODUCTION

Areas of the Qu'Appelle River system, located in southern Saskatchewan, have been found to be contaminated with mercury. High concentration of mercury were measured in sediments from the Moose Jaw River (0.51-1.5 ppm Dry Weight), Wascana Creek (2.0-5.4 ppm) (Hammer *et al.* 1982) and Thunder Creek (0.1-38 ppm) (Gummer 1980). Aquatic biota from the Qu'Appelle system have also accumulated mercury (Hammer *et al.* 1982; Munro and Gummer 1980).

A number of factors influence the partitioning of mercury between the sediments, water and biota of aquatic ecosystems. Aquatic sediments contain the major portion of the mercury present in aquatic ecosystems. The amount of mercury accumulated by the sediments has been related to its organic content and mean grain size (Jackson 1979; Ramamoorthy and Rust 1978). Mercury levels in water are affected by the amount of organic matter and particulate material in the water (Ottawa River Project Group 1979). The chloride content of water also influences the mercury level in water (Feick *et al.* 1972; Ramamoorthy and Rust 1978).

The lakes in the Qu'Appelle system are subsaline (Hammer 1983), with average chloride levels ranging in 1977 from 12 ppm in Buffalo Pound Lake to 75 ppm in Katepwa Lake (Inland Waters Directorate 1982). How different chloride levels affect the uptake and release of mercury from a Qu'Appelle lake sediment (Buffalo Pound) and from a sand substrate was the first objective of this study. The second objective was to examine the subsequent bioaccumulation of mercury by some aquatic organisms: amphipods (*Hyalella azteca*) and rainbow trout (*Salmo gairdneri*).

MATERIALS AND METHODS

Three experiments were conducted. Sediments from Buffalo Pound Lake were used for the first two experiments, while sand was used in the third experiment. The Buffalo Pound Lake sediments were obtained on May 31, 1984 from depths of 3-5m off Valleyview Beach. Total and volatile residues (APHA 1980) were determined for the sediments and sand before each experiment. Volatile residue is a maximum estimate of the organic carbon content and also includes volatile inorganic salts (APHA 1980). Differences in the total residues (Table 1) between experiments I and II are attributed to drying of the sediments during storage.

The first experiment exposed the amphipod, Hyaella azteca, to a control and three treatments. The control had no mercury added while treatments 1, 2 and 3 received 550 ug of HgCl_2 (400 ug Hg) dissolved in distilled water at Day 0. Chloride levels are given in Table 1. Seven replicates were used for the control and each treatment.

A two litre Pyrex glass beaker was used for each replicate. Each beaker was previously exposed to a 100 ppb Hg solution for 8 days, so that any adsorption sites on the inner glass surfaces of the beakers would be occupied by mercury.

Each replicate had approximately 139 g (Dry Weight) of sediment. Plants (Myriophyllum sp.) (10-20 g Wet Weight) were added to each replicate but most did not survive. Dechlorinated tap water (Total Hardness 380 ppm) was used in each replicate. On Day 10 approximately 70 amphipods were added to five of the replicates in the control and treatments. The remaining two replicates were removed and sampled on Day 11 for total mercury in the sediments and water. Two more replicates were sampled on Day 25 for mercury in the water and amphipods and the final three replicates were sampled on Day 44 for mercury in the sediments, water and amphipods. Initial levels of mercury in the amphipods were established from three subsamples taken when the amphipods were added to the replicates.

Water samples (250-300ml) for all experiments were preserved by adding 1.7 mL concentrated HNO_3 to each sample. Samples were stored at 4.0°C in the dark and were analysed within 28 days of the sampling date. Polypropylene sample containers were cleaned by rinsing them with 1N HCl followed by 1N HNO_3 and finally with distilled water. Sediment samples were kept frozen until analyzed. Amphipods were frozen, freeze dried and ground to a fine powder before analysis.

Chloride levels in water were analyzed in all experiments using the Mohr method (HACH 1984). Temperature was monitored from Monday to Friday, oxygen 2-3 times a week and pH twice a week in each experiment (Table 1). A YSI Model 54 meter and probe were used to measure oxygen and Chemtrix Type 40 E meter and combination probe were used to measure pH.

The second and third experiments exposed rainbow trout to similar chloride levels (Table 1) and total amounts of mercury added, but over different substrates. Both experiments had a control and two treatments, each with two replicates. Approximately, 1490 g and 6100 g (Dry Weight) of sediment and sand, respectively, were placed in the replicates of Experiment II (sediment) and Experiment III (sand). Mercuric chloride ($6100 \text{ ug } \text{HgCl}_2$, 4500 ug Hg) was

dissolved in distilled water and added to each replicate of treatments 1 and 2 in both experiments. If all the mercury was taken up by the sediments and sand, the resulting mercury concentration would be 3.1 ppm and 0.7 ppm in the sediment and sand respectively.

Glass aquaria (75 x 30 x 37.5 cm) were used for each replicate in Experiments II and III. There was no pretreatment of the aquaria with a mercury solution. A fiberglass screen (mesh 1.6 x 1.6 mm) was placed 8-10 cm above the sediments in Experiment II to separate the fish from the sediments. No screen was used in Experiment III. Tap water was added to each replicate in both experiments.

Chloride levels were increased by adding NaCl to the respective treatments in Experiments I, II and III.

Nine (9.8-17.4 cm Fork Length) and six (16.1-26.8 cm FL) trout were added on Day 7 to each replicate in Experiments II and III, respectively. The fish were anaesthetized with MS 222 and measured for fork length and wet weight (g), and marked by fin clips. Initial levels of mercury in the trout were established in Experiments II and III by sacrificing 4 fish when the trout were added to the replicates on Day 7.

Water and sediment samples were taken for mercury analyses on Day 7, 21 and 35 in Experiment II and on Days 1, 4, 7, 14, 21, 28 and 35 in Experiment III. Fish were sampled on Days 21 and 35 in both experiments. After sacrificing, sections of the white muscle were removed from each fish. These sections were freeze dried prior to mercury analyses.

Fish were fed 3-4 times a week in Experiment II and 4-5 times a week in Experiment III. Trout Grower Finisher pellets were used as food. Analysis of these pellets showed a mercury content of 0.02-0.05 ppm (Dry Weight).

Mercury analyses of water, sediment, amphipod and trout samples were carried out by the Saskatchewan Research Council according to methods outlined by Environment Canada (1977).

Analysis of variance and Duncan's Multiple Range Test (Steel and Torrie 1960) were used to compare the mercury concentrations of water, sediment, amphipod and trout samples between treatments and with the controls of the respective experiments.

RESULTS AND DISCUSSION

Except for the fluctuating mercury levels in the water

of treatments 1 and 2 in Experiment II (Fig. 1), an equilibrium level was established in 10-14 days in all the controls and treatments (Fig. 2 and 3). The decrease from high initial mercury levels in the water was significant (Expt. I: $p = 0.01$, F Value = 15.09, df 2: 22) (Expt. III: $p = 0.01$, F Value = 10.231, df 6: 33) over time for the various treatments in Experiments I and III. The equilibrium level was 0.2-0.6 ppb regardless of whether mercury had or had not been added to the system. The exception was treatment 2 in Experiment III where the mercury equilibrium level was 2.0 ppb (Fig. 3).

These equilibrium levels are similar to or slightly higher than those observed in water in field studies. Jackson *et al.* (1982) reported total particulate and total dissolved mercury levels of 0.05-0.37 ppb and 0.01-0.13 ppb, respectively, in the mercury polluted Wabigoon River system. Mercury levels in the Qu'Appelle system have ranged from 0.13 ppb (total Mercury) in the Moose Jaw River (Gummer 1980) to between 0.02-0.09 ppb (Extractable Mercury) for Buffalo Pound Lake (Inland Waters Directorate 1982).

Fluctuating levels of mercury in water, seen in treatments 1 and 2 of Experiment II (Fig. 1) are attributed to the disturbance of the sediments by the trout. Trout got underneath the screens in at least 1 replicate of the control and each treatment and disturbed the sediments. This resulted in large amounts of silt being suspended in the water. Elevated but variable mercury levels occurred in treatments 1 and 2. Gummer (1980) observed that maximum mercury levels in the water of the Moose Jaw River coincided with maxima in water flow levels, suspended sediment and particulate organic matter. The level of mercury in the water is likely to increase when mercury contaminated sediments become suspended in the water. The sand sediments used in Experiment III were also disturbed by the trout but the sand did not stay suspended. This is attributed to the larger particles size of the sand.

Higher mercury equilibrium levels in the water of treatment 2 (Experiment III) (Fig. 3) result from the increased chloride levels in the water. Feick *et al.* (1972) observed that the addition of NaCl or CaCl₂ increased the relative amount of mercury in water in equilibrium with sand or organic sediments by 2 to 5 orders of magnitude. However, our experiments show that the increased chloride only resulted in elevated mercury levels in water over sand. The difference in the results of the two studies lies in the fact that in the experiments of Feick *et al.* (1972), the mercury levels in the sediments were 400 to 2,000X greater and the chloride levels were 125X greater. They stated that the chloride effect tends to increase as the mercury burden of the sediment increases. Ramamoorthy and Rust (1978) observed that two Ottawa River sediment samples with mercury

levels of 4.1 and 11.0 ppm desorbed 71 and 78%, respectively, of the mercury in the presence of a 10^{-4} M NaCl solution (3.6 ppm Cl⁻) over a 4-day period. The difference between these latter results and our own are caused by differences in the characteristics of the sediments used in the respective studies. The interaction of sediment characteristics with chloride and their effect on mercury adsorption and desorption by sediments is discussed later.

The majority of the mercury that remained in the systems of the respective experiments was in the sediments. In Experiment I almost 75% (Table 2) of the mercury added was in the sediments in treatment 1 while treatments 2 and 3 retained smaller amounts. The latter two treatments had higher chloride levels in the water. This suggests that the chloride affected the adsorption of the mercury by the Buffalo Pound Lake sediments. However, in Experiment II the reverse is true in the amount of mercury retained by Buffalo Pound Lake sediments exposed to waters with different chloride levels (Table 3). These sediments were disturbed by the trout and the suspension of particulate and organic matter may have affected the amount of mercury retained. The percentage of mercury retained by the sand in both treatments of Experiment III was considerably lower (Table 4) than percentages retained in either Experiment I (Table 2) or II (Table 3). The Buffalo Pound Lake sediments appear to be able to adsorb more mercury than the sand. The mercury concentration in the sediments of all treatments in the experiments was higher than the controls (Fig. 4, 5 and 6). However, only in Experiments I and III were the concentrations significantly (Expt. I: $p = 0.01$; F Value = 15.840; df 3: 15) (Expt. III: $p = 0.05$; F value = 3.478; df 2:28) different. Also the mercury sediment concentrations of treatments 2 and 3 in Experiment I did not differ significantly from treatment 1. The lack of a significant difference between treatments 1, 2 and 3 (Experiment I) and between the control and treatments of Experiment II is thought to result from a small sample size.

The lost mercury that was not taken up by the biota and sediments or that remained in the water was likely volatilized. Reimers *et al.* (1974) notes that mercury is known for its high volatility.

It has been observed that the majority of mercury that enters aquatic ecosystems is taken up by the sediments. Kudo *et al.* (1977) estimated that 97% of the total mercury present in the Ottawa River was in the sediments. Rogers *et al.* (1981) concluded that the high sorption capacity of sediments for mercury and the relatively very slow desorption resulted in the majority of the mercury in aquatic ecosystems being in the sediments. Hammer *et al.* (1982) found a highly significant correlation between the

total mercury content and the percentage of organic carbon in the sediment of the Qu'Appelle River system. Jackson (1979) found a similar correlation in the sediment from the Wabigoon River system. The partition coefficient of mercury between sediments and water from the Ottawa River increased with decreasing particle size and increasing organic content of the sediment and decreased with increasing particulate and organic material in water (Ottawa River Project Group 1979).

The Buffalo Pound Lake sediments differed substantially from the sand sediment used in Experiment III and from the sediments used by Ramamoorthy and Rust (1978) in percent organic carbon and particle size. Buffalo Pound Lake sediment had a larger percentage of organic content (3.6%) and the majority of the sediment particles were quite small ($60.1\% < 5 \mu\text{m}$) (Oscarson *et al.* 1981). The two sediments used by Ramamoorthy and Rust (1978) had an organic content and average particle size of 0.6% and $267 \mu\text{m}$ respectively for one sediment and 2.4% and $151 \mu\text{m}$ for the second. The sand sediment of Experiment III had very large particles ($83.3\% > 500 \mu\text{m}$) and a volatile residue of 0.9%.

Buffalo Pound Lake sediments with their greater organic content and very small particle size are not affected by a water chloride content of up to 170-180 ppm in the equilibrium level of mercury in the water. The sediments do appear to be affected by chloride in the amount of mercury adsorbed by the sediments. The sand sediments do not adsorb mercury very readily at both low and high chloride concentrations. The high chloride concentration in water over sand resulted in higher mercury equilibrium levels.

The amount of mercury in the amphipods is difficult to estimate since many of the samples from the control and treatments had mercury below detection limits. The latter varied considerably depending on the sample weight available for analysis. The minimum amount of mercury that can be detected in a sample depends on the size of the sample. The minimum is 0.05 μg of mercury in 0.5 g (Wet Weight) of sample (W. Yuen, Pers. Comm.). When the sample size is less than 0.5 g there must be more mercury present in the sample before it can be detected.

Only one replicate from each of treatments 1, 2 and 3 on Day 25 had concentrations of mercury at detectable levels, 4.8 ppm, 2.9 ppm and 8.4 ppm, respectively (Table 5). These values are higher than the mercury levels established for the three initial samples of amphipods on Day 10. This indicates that there was accumulation of mercury by the amphipods. However, the amount accumulated is quite variable within and between treatments and is not related to the chloride content of the water. Mercury accumulation by the amphipods was quite variable over time. Mercury levels

for Day 44 (Table 5) were quite variable and were often less than the levels detected on Day 25. Variations over the year have been seen in the mercury concentrations in amphipods. Kristenson (1982) observed that the mercury concentration of Gammarus pulex varied from 1.01 to 7.08 ppm (Dry Weight). Peak values occurred in June while significantly lower values occurred in May and July. He suggests this maximum in mercury concentration results from an optimizing of conditions for methylation of mercury by micro-organisms. Zauke (1977) observed decreases in mercury concentration at some sample sites for 3 species of Gammarus from spring and summer to late autumn along the Elbe River. At other sites no seasonal trends were evident. However, the range of variation in mercury levels at individual sites on the same date was not as large as was seen in our experiments. Zauke (1977) suggested that part of the seasonal loss of mercury may occur through moulting of the exoskeleton.

In both of our experiments with trout there was a significant (Expt. II: $p = 0.01$; F Value = 33.47; df 3: 24) (Expt. III: $p = 0.01$; F Value = 9.304; df 1: 27) increase over time in the mercury level of the trout (Fig. 7 and 8). This was especially evident in the treatments (1 and 2) in which mercury had been added. However, only in Experiment I was the increase in mercury concentration of the trout great enough to be significantly (Expt. II: $p = 0.01$; F Value = 47.62; df 2: 50) (Expt. III: $p = 0.05$; F Value = 1.286; df 2: 27) different from the controls. A second difference in the uptake of mercury by the trout was that in Experiment II the increased levels of mercury were reached within the first 14 days, but in Experiment III the mercury content of the trout only increased noticeably during the second 14 day period (Fig. 7 and 8).

There are two possible pathways for mercury uptake by fish. The first is by release of mercury from the sediments to the water followed by uptake through the gills. The second is from the food through the gastro-intestinal tract. In our experiment mercury uptake is mainly through the first pathway, as the food had a very low mercury content. Olson *et al.* (1973) suggests that the uptake of Hg^{2+} and methyl mercury (CH_3Hg^+) occurs mainly through the gills, with the latter form being taken up more readily than the former.

Studies have shown that the organic matter content of aquatic sediments will influence the methylation of mercury (Jackson and Woychuk 1981; Akagi *et al.* 1979), with more methyl mercury usually produced with increasing organic content of sediments. Langley (1973) reported that the highest methylation rate did not coincide with the highest mercury concentration in the sediment but rather with organically enriched sediments with high microbial activity.

We suggest that the delay in mercury uptake in Experiment III may be the result of the slow establishment of a microbial population in the sand. This population would be needed to methylate the mercury which would then be more readily taken up by the fish. The Buffalo Pound Lake sediments would already have an established microbial population, thus there would be no lag in mercury methylation. The sand would not have a resident microbial population and it would take time to establish one. Langley (1973) observed a lag period of less than 1 week in the rate of methylation of mercury by lake sediment microbial populations. This lag period was thought to result from the inactivation of methanogenic bacteria. The inactivation was caused by disturbance of the sediments during the setting up of the experiment (Langley 1973). Jensen and Jernelov (1969) found that methylation of HgCl_2 did not occur in sterilized sediments. This shows the importance of microbial populations for methylation of mercury.

Why there was no further increase in mercury levels in the trout during the second 14 day period in Experiment II is not clear. It may be that the fish reach an equilibrium level of mercury, where the amount eliminated is equal to the amount taken up.

Another question is why there was no significant difference in mercury uptake by trout between treatments 1 and 2 of Experiment III, even though the latter treatment had a higher equilibrium concentration of mercury in the water. As stated above methyl mercury is taken up more readily through the gills than Hg^{2+} . It may be that the increased mercury levels in the water were mainly in the form of inorganic mercury (Hg^{2+}). This inorganic mercury was not taken up very readily by the fish.

The mercury levels of the individual fish removed from the same replicate on the same date were quite variable. Similar results have been observed by Huckabee *et al.* (1975) and Wobeser *et al.* (1970). This indicates that individual fish vary in their ability to take up and eliminate mercury. The mercury content of fish was not related to the size of the fish.

ACKNOWLEDGEMENT

Dennis Dyck prepared the figures for this manuscript. The research was supported by NSERC Strategic Grant No. G1296.

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Table 1. Data for temperature, oxygen, chloride, and pH in water in Experiment I. (Control, no Hg added, Treatments 1, 2, 3 400 ugHg added); Experiment II (Control, no Hg added, Treatment 1, 2 4500 ugHg added); and Experiment III (Control, no Hg added, Treatment 1, 2 4500 ugHg added). Total and volatile percentages of original wet weight of sediments.

Experiment I Buffalo Pound Lake				
Sediment Treatment	Control	1	2	3
Temperature (°C)	17.3-18.9	17.4-18.9	17.5-19.5	17.5-19.7
Oxygen (ppm)	7.9-8.7	7.4-8.6	8.0-8.6	7.8-8.7
pH	8.30-8.65	8.30-8.65	8.30-8.65	8.30-8.65
Chloride (ppm)	24	26	74	160
Total Residue (%)	32.2			
Volatile Residue (%)	8.0			

Experiment II Buffalo Pound Lake			
Sediment Treatment	Control	1	2
Temperature (°C)	16.1-17.5	15.5-17.5	15.2-17.3
Oxygen (ppm)	8.5-9.2	7.7-9.4	7.5-9.0
pH	8.00	8.02	8.00
Chloride (ppm)	16-18	13-17	166-184
Total Residue (%)	42.4		
Volatile Residue (%)	7.2		

Experiment III Sand			
Sediment Treatment	Control	1	2
Temperature (°C)	15.2-15.8	14.5-15.0	14.2-15.2
Oxygen (ppm)	8.6-8.9	8.9-9.6	9.1-9.7
pH	7.60-8.20	7.60-8.20	7.60-8.20
Chloride (ppm)	10-14	9-14	167-175
Total Residue (%)	84.1		
Volatile Residue (%)	0.9		

Table 2. Total mercury content (ug) on day 44 in water and sediment compartments. Experiment I. (Total mercury content of sediment = Conc. of Hg (ug.g^{-1}) $\times$ 139g sediment 139g, Dry Wt. Total mercury content of water = Conc. of Hg(ug.L^{-1}) $\times$ 1.7L)

Treatment	Control	1	2	3
Hg added (ug)		400	400	400
Cl ⁻ (ppm)	24	26	74	160
Water (ugHg)	0.49	0.66	0.63	0.60
Sediment (ugHg)	6.04.	304.92	166.32	148.30
Total (ugHg)	6.53	305.58	166.95	148.90
Percent Hg Remaining		74.8	40.1	35.6

Table 3. Total mercury content (ug) on day 35 in water and sediment compartments. Experiment II. (Total mercury content of sediment = Conc. of Hg(ug.g^{-1}) $\times$ sediment 1492g Dry Wt. Total mercury content of water = Conc. of Hg(ug.L^{-1}) $\times$ 76.7L)

Treatment	Control	1	2
Hg Added (ug)		4500	4500
Cl ⁻ (ppm)	16-18	13-17	166-184
Water (ugHg)	20.71	145.7	383.5
Sediment (ugHg)	89.52	1163.8	2611.0
Total (ugHg)	110.23	1309.5	2994.5
Percent Hg Remaining		26.7	64.1

Table 4. Total mercury content (ug) on day 35 in water and sediment compartments. Experiment III. (Total mercury content of sediment = Conc. Hg (ug.g⁻¹) x sediment 6,053g Dry Wt. Total mercury content of water = Conc. Hg (ug.L⁻¹) x 76.7L)

Treatment	Control	1	2
Hg added (ug)		4500	4500
Cl ⁻ (ppm)	10-14	9-14	167-175
Water (ugHg)	32.21	46.0	149.6
Sediment (ugHg)	163.40	647.7	617.4
Total (ugHg)	195.61	693.7	767.0
Percent Hg Remaining		11.1	12.7

Table 5. Estimated levels of total mercury in the amphipods (Hyaletia azteca) (ppm Dry Wt) over time. (< denotes that mercury concentration is less than the accompanying number.)

Treatment	Control	1	2	3
Hg added (ug)		400	400	400
Cl ⁻ (ppm)	24	26	74	160
Day 10	< 2.1	< 2.0	< 1.3	
25	< 1.7	4.8	2.9	8.4
	< 3.8	< 2.1	< 2.6	< 3.4
44	< 10	< 3.5	< 3.2	< 4.4
	< 5.6	< 3.7	< 5.8	< 7.2
		< 6.0	< 7.7	< 7.9

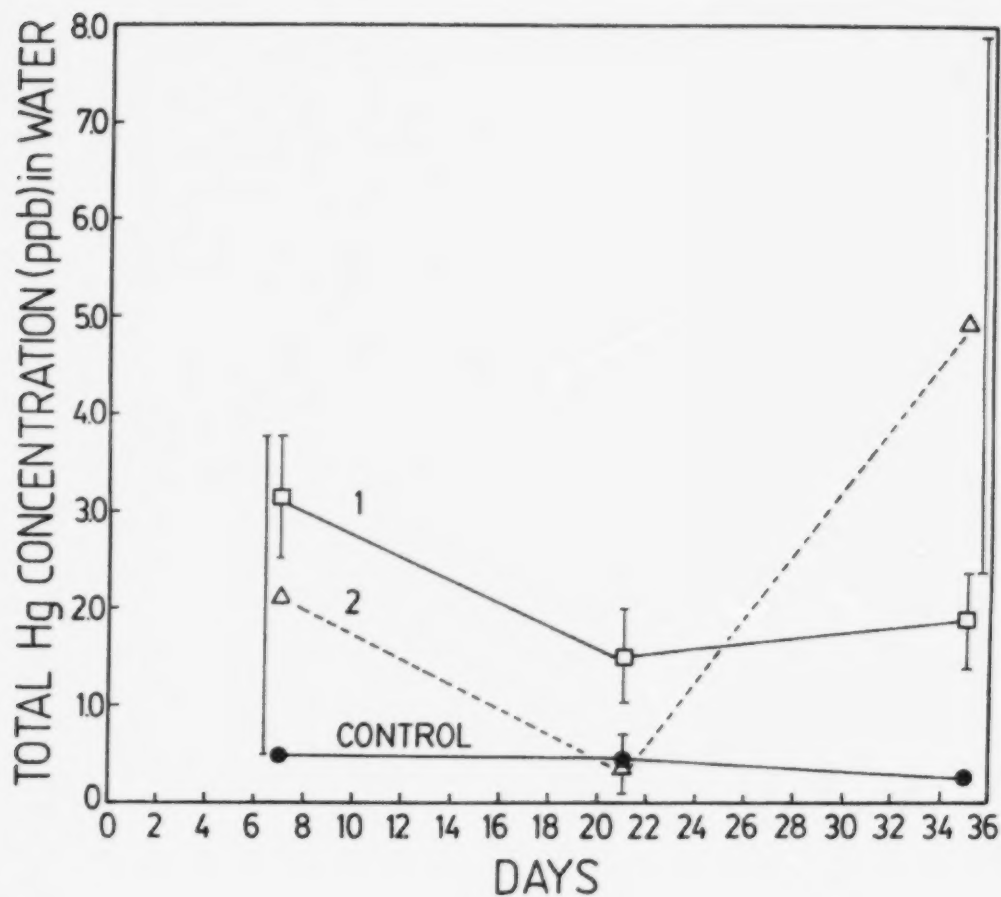


FIG. 1. Temporal concentration of Total Mercury (ppb) in water over Buffalo Pound Lake sediments for Control (No Hg Added; Cl^- Range 16-18 ppm), Treatment 1 (Hg Added 4500 ug; Cl^- Range 13-17 ppm) and Treatment 2 (Hg Added 4500 ug; Cl^- Range 166-184 ppm).

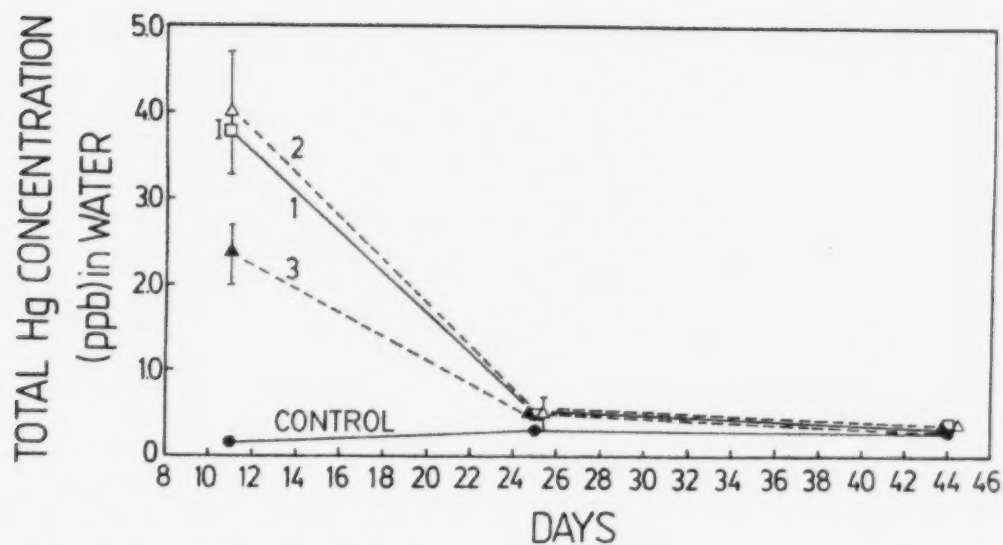


FIG. 2. Temporal concentration of Total Mercury (ppb) in water over Buffalo Pound Lake sediments for Control (No Hg Added; Cl⁻ 24 ppm), Treatment 1 (Hg Added 400 ug; Cl⁻ 26 ppm), Treatment 2 (Hg Added 400 ug; Cl⁻ 74 ppm) and Treatment 3 (Hg Added 400 ug; Cl⁻ 160 ppm).

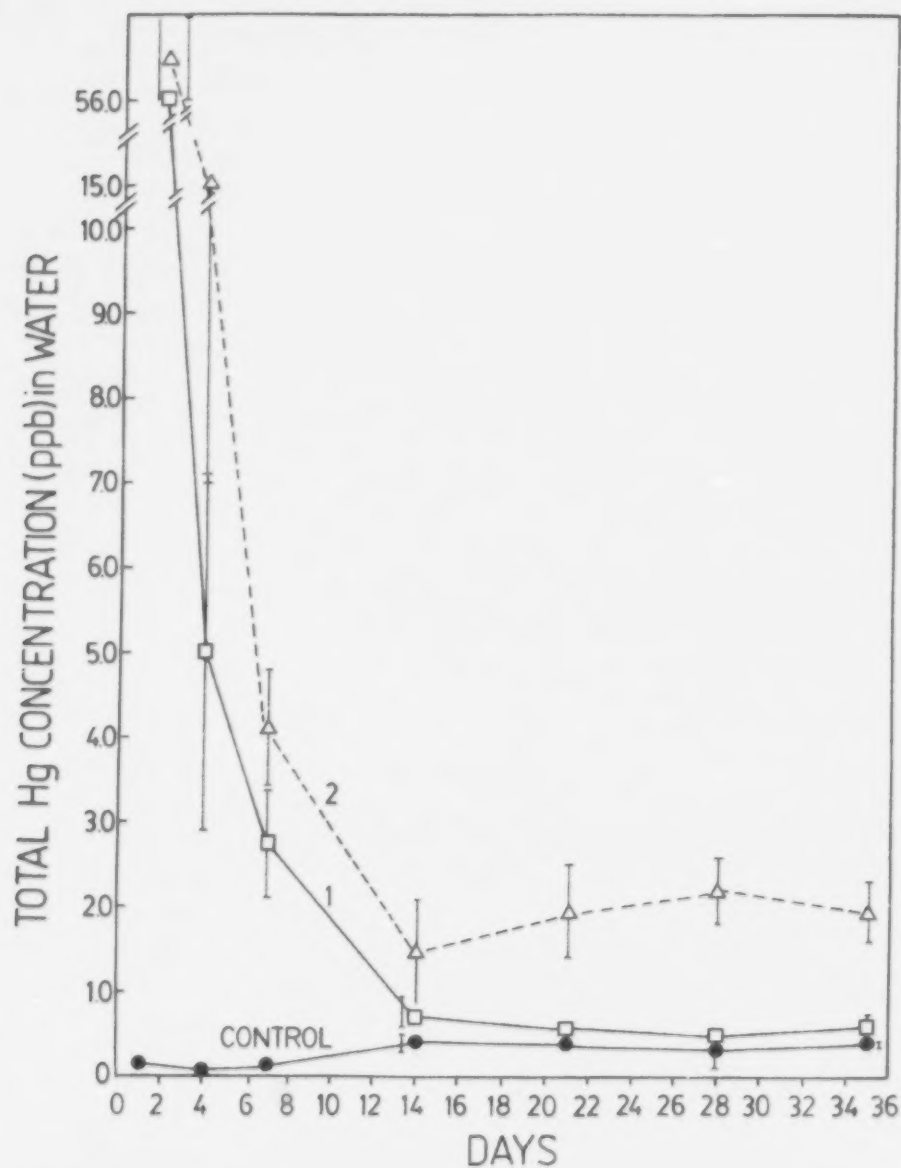


FIG. 3. Temporal concentration of Total Mercury (ppb) in water over sand sediment for Control (No Hg Added; CI- Range 10-14 ppm), Treatment 1 (Hg Added 4500 ug; CI- Range 9-14 ppm), Treatment 2 (Hg Added 4500 ug; CI- Range 167-175 ppm).

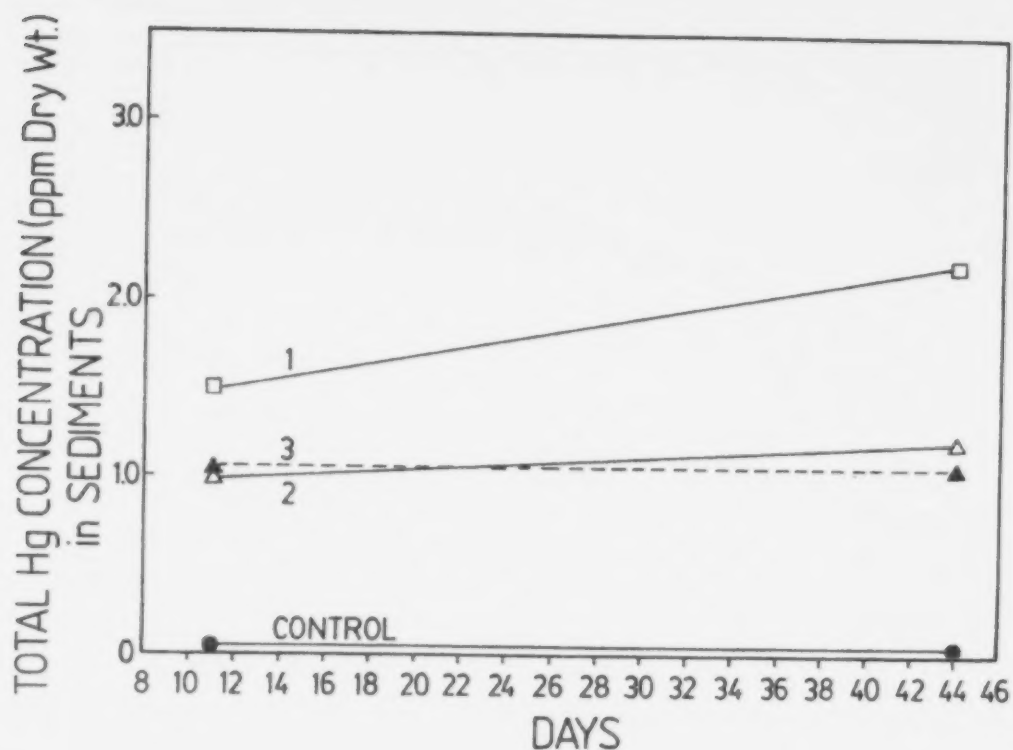


FIG. 4. Temporal concentration of Total Mercury (ppm Dry Weight Sediment) in Buffalo Pound Lake sediments for Control (No Hg Added; Cl^- 24 ppm), Treatment 1 (Hg Added 400 μg ; Cl^- 26 ppm), Treatment 2 (Hg Added 400 μg ; Cl^- 74 ppm) and Treatment 3 (Hg Added 400 μg ; Cl^- 160 ppm). Average of two replicates (Day 11) and three replicates (Day 44).

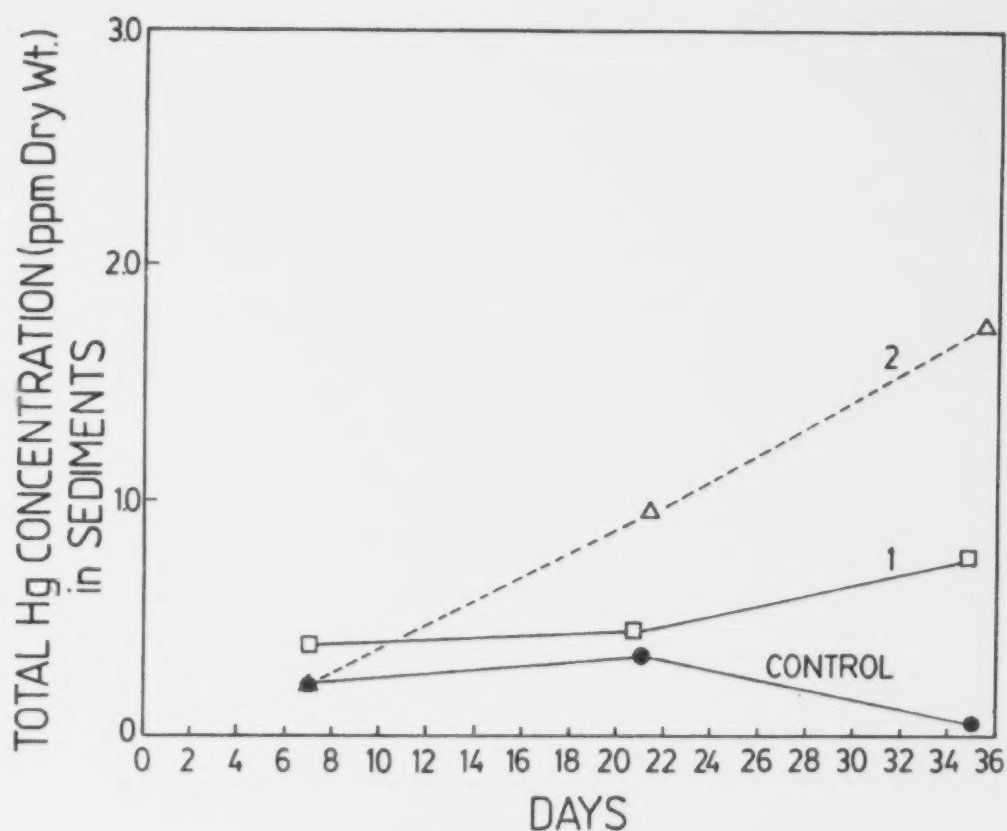


FIG. 5. Temporal concentration of Total Mercury (ppm Dry Weight Sediment) in Buffalo Pound Lake sediments for Control (No Hg Added; CI-Range 16-18 ppm), Treatment 1 (Hg Added 4500 ug; CI-Range 13-17 ppm) and Treatment 2 (Hg Added 4500 ug; CI-Range 166-184 ppm). Average of two replicates.

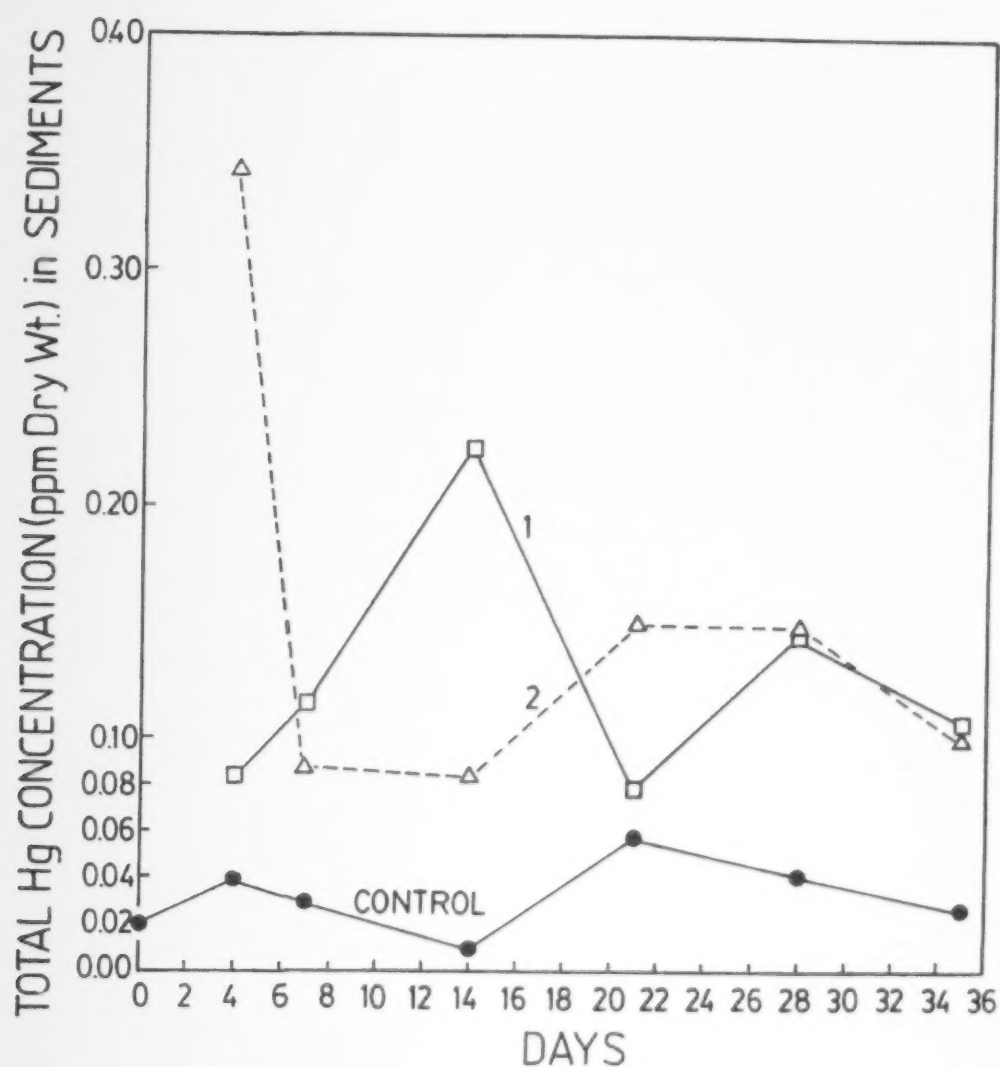


FIG. 6. Temporal concentration of Total Mercury (ppm Dry Weight Sediment) in sand sediments for Control (No Hg Added; Cl⁻ Range 10-14 ppm), Treatment 1 (Hg Added 4500 ug; Cl⁻ Range 9-14 ppm) and Treatment 2 (Hg Added 4500 ug; Cl⁻ Range 167-175 ppm). Average of two replicates.

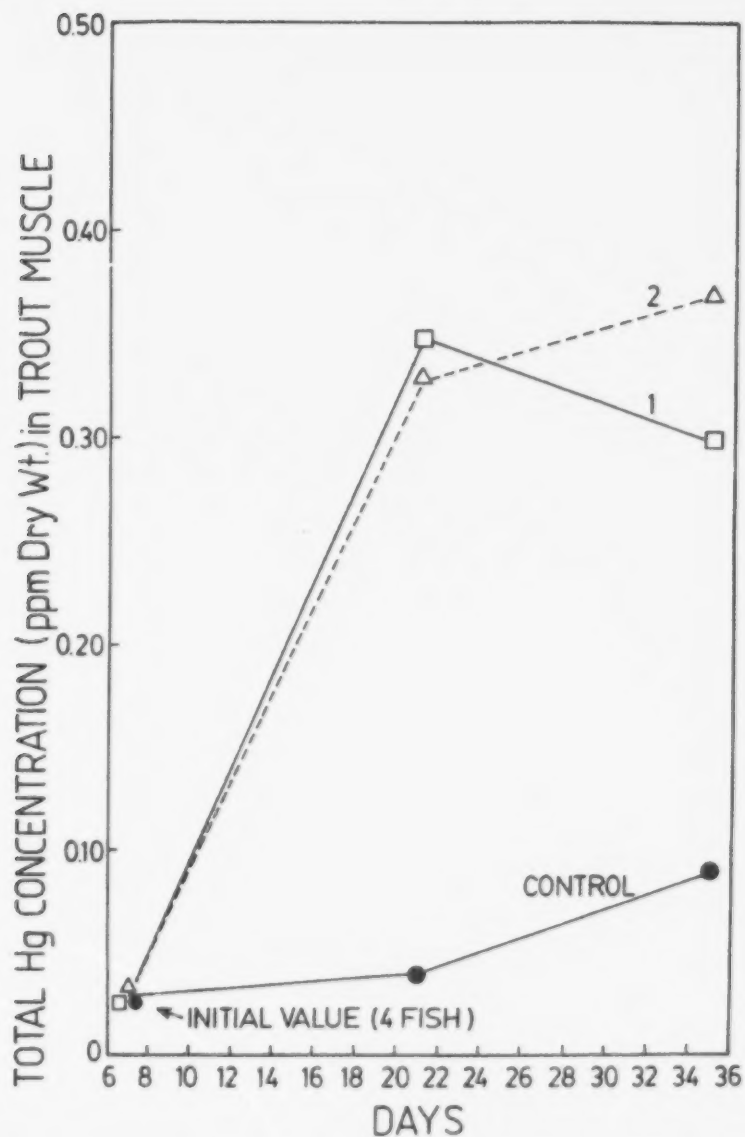


FIG. 7. Temporal concentration of Total Mercury (ppm Dry Weight) in trout muscle for Control (No Hg Added; Cl⁻ Range 16-18 ppm), Treatment 1 (Hg Added 4500 ug; Cl⁻ Range 13-17 ppm) and Treatment 2 (Hg Added 4500 ug; Cl⁻ Range 166-184 ppm). Buffalo Pound Lake sediment. Average of eight fish.

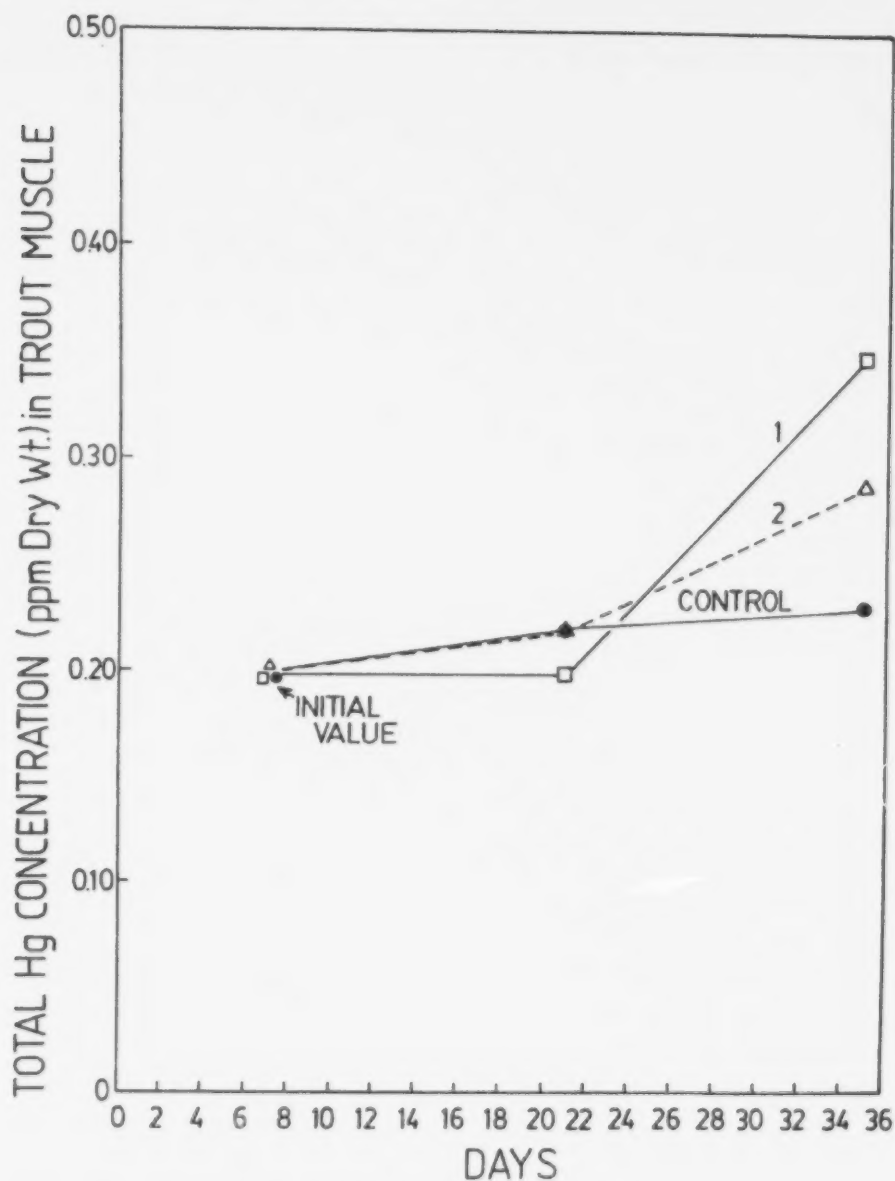


FIG. 8. Temporal concentration of Total Mercury (ppm Dry Weight) in trout muscle for Control (No Hg Added; Cl⁻ 10-14 ppm), Treatment 1 (Hg Added 4500 ug; Cl⁻ Range 9-14 ppm) and treatment 2 (Hg Added 4500 ug; Cl⁻ Range 167-175 ppm). Sand sediment. Average of six fish.

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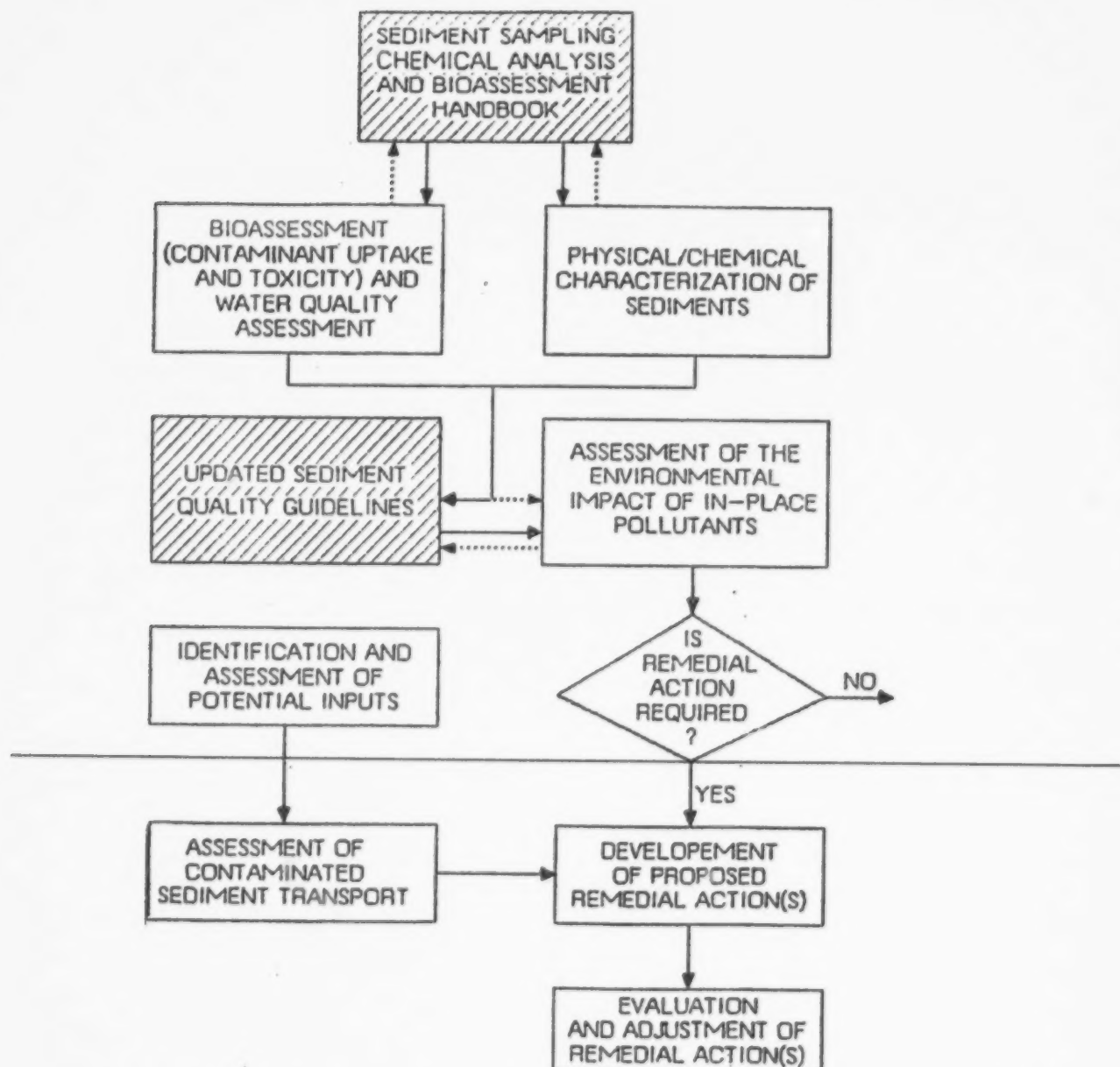
In-Place Pollutant Working Group

Sediments are a major repository of toxic contaminants entering the aquatic environment. As contaminants enter the Great Lakes many of them adhere to fine particles and eventually sink to the bottom of lakes. Once settled, these contaminant laden sediments create an in-place pollutant problem posing a long-term threat to the aquatic ecosystem. These sediments may be resuspended through human activities (eg. dredging and ship traffic) or natural processes, such as storms, providing a source of contaminants to the water column and aquatic biota. Even undisturbed contaminated bottom sediments are potentially harmful since they may be absorbed by benthic organisms causing bioaccumulation within the ecosystem.

The need to assess the environmental impacts of contaminated bottom sediments in the Great Lakes has become apparent in the last few years. The gradual build up of contaminants in localized areas has resulted in many river mouths and harbours being characterized as "Areas of Concern". In 1983, the International Joint Commission's Water Quality Board identified 38 of the 42 Areas of Concern as having major in-place pollutant problems.

In recognition of the severity of in-place pollutant problems in the Great Lakes and the significant number of Areas of Concern with contaminated sediments, a joint Ministry of Environment (MOE)-Environmental Protection Service (EPS) Technical Working Group has been formulated to assess problem areas on the Canadian Great Lakes.

It is hoped that the In-Place Pollutants Working Group can contribute directly or indirectly to the development of Remedial Action Plans required by the Water Quality Board. The Working Group will provide scientific and technical advice and support in the formulation



NOTE. While under development, the "Sampling Handbook" and "Updated Sediment Quality Guidelines" will represent programme outputs, ultimately, however, they will be used as inputs.

Figure 1

and implementation of those Remedial Action Plans where in-place pollutant problems are identified as being significant enough to warrant remedial measures. The Working Group will focus on the development of methodologies for the assessment of specific in-place pollutant problems, as well as the evaluation of remedial action alternatives.

To date The Working Group has initiated a review of MOE's Bulk Chemical Criteria for Open Water disposal. Preliminary findings demonstrate the inadequacy of using just bulk chemical criteria to characterize contaminant levels in sediment. Consequently a methodology and supporting rationale is being formulated for assessing various levels of acceptability for heavy metals and organic contaminants based upon both bulk chemical characterization of sediment and the bioavailability and bioaccumulation potential of contaminants (see figure 1).

In order to facilitate contaminant characterization the Working Group has initiated the development of field and laboratory protocols to be used in evaluating in-place pollutant problems. These protocols will be used in establishing the physical and chemical characteristics of sediment, the influence of the physical characteristics and chemical quality of sediments on the distribution and abundance of benthic organisms and the role of contaminants in sediments on benthic bioaccumulation and water quality impairment.

The first field verification of these protocols involves a series of sediment bioassay experiments, undertaken to evaluate the short-term and long-term impacts of contaminated dredge spoils disposal. It includes both acute toxicity and bioaccumulation studies. A preliminary experiment has been run to evaluate bioassessment methods used by the U.S. Environmental Protection Agency and the U.S. Fish and Wildlife Service (with modifications we felt were necessary to improve the experimental design). Two methods were used to assess the long-term impact of dredged spoils disposal. Both utilized test sediments covered with water. The major difference between the two was that one, the Prater-Anderson apparatus, consisted of a recirculating system while the Beaker tests did not. The experiments to be carried out in our study are designed to assess the reproducibility of the tests and to compare the two methods.

The concerns associated with contaminated sediments relate to the bioaccumulation and toxicity to organisms in, on and above the sediments. The organisms selected therefore cover all of these concerns:

	In Sediment	On Sediment	In Water
Toxicity	Hexagenia Limbata	Hyallorella Azteca	Pinephales Promeles (Fathead minnows)
Bioaccumulation	Lumbricus terrestrius (earth worm)	Hyallorella Azteca	Pinephales Promeles

TABLE 1

The sediments used in this study were from the Spadina Slip in Toronto Harbour. They were contaminated with both heavy metals and PCB's (see Table 2). Control sediments were taken from Georgian Bay.

The preliminary results demonstrated that the methodologies were very sensitive for toxicity demonstrating a significant mortality rate associated with the contaminated sediments. With regard to bioaccumulation, however, there are two possible conclusions:

1. The contaminants in the sediments tested are not readily bioavailable;
2. The test methods are not sensitive.

Some initial problems were encountered with the Prater-Anderson Apparatus, however, with some modifications it was felt that it was the preferred over the Beaker methodology.

	MOE Guidelines for Open Water Spoil Disposal	Spadina Slip (Contaminated Sediments)	Georgian Bay (Control)
Arsenic	8.0	10.5	4.5
Cadmium	1.0	4.9	0.3
Chromium	25	78	66
Copper	25	230	21
Iron	10,000	24,500	51,600
Lead	50	780	27
Mercury	.3	1.29	0.10
Zinc	100	850	195
PCB (ug/kg)	50	110	24

Figure 3

* All values unless otherwise noted are shown as mg/kg dry weight basis.

A second series of experiments has been initiated using a range of contaminated materials. The results of these are not yet available.

Future experiments will focus on evaluating the toxicity of sediments high in PCB's without elevated metal levels.

After protocols have been established, the Working Group will use these substantiated methodologies together with the findings from the Bulk Chemical Criteria Review to undertake case-by-case evaluations of insitu contaminant problems.

The Working Group will have developed a comprehensive methodology for characterizing contaminated sediment problems in Areas of Concern and will therefore have valuable input into the development of Remedial Action Plans.

HAZARD ASSESSMENT OF CHEMICALS IN THE AQUATIC ENVIRONMENT

Dr. W.M.J. Strachan

Abstract

There are at least 963 chemicals identified as present in the Great Lakes. There are undoubtedly others in other locations and in these waters. The problem or question is - what are their significances in the environments. There have been a number of attempts to describe processes to evaluate the hazard which chemicals represent. These have been undertaken for a variety of purposes and hence have taken account of an equally varied list of chemical characteristics. Some assessment procedures have attempted to reduce the process to a formula; others have relied on panels of experts.

The International Joint Commission and Environment Canada, separately, are interested in undertaking hazard assessment of the Great Lakes chemicals. The purpose would be to screen and prioritize the chemicals for specified data development with a view to subsequent hazard evaluation and possible regulation. Unfortunately, there is a paucity of even the most rudimentary information on most of the chemicals although there are ways of predicting much of the data; a concern is the reliability of assessments based on such estimations.

Two particular questions are directed at the Aquatic Toxicity Workshop attendees - what are the minimum data elements which should be evaluated for screening based on hazard to/from the aquatic environment, and, in what manner should the individual elements be combined to produce acceptable expressions for exposure and effects. A small workshop is being undertaken in late November and attendees are asked to respond prior to this to:

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Applicability of Laboratory Studies for Predicting Field Events -

Introduction

D. I. Mount

Laboratory studies are done to predict field events of many types. When the purpose relates to toxicity concerns, toxicity tests are often used. Toxicity tests measure species sensitivity - nothing else. Therefore to predict community effects from such tests, only those effects that are directly caused by sensitivity to chemicals are valid for comparisons. Toxicity stresses the more sensitive species. It does not "unstress" the more tolerant ones! However more tolerant species may change in numbers as a result of more sensitive species being decreased. Such effects cannot be predicted with toxicity tests.

Field studies do not measure only changes from toxicity stress but changes due to all causes. Rarely can field measurements prove the cause of changes. Toxicity tests can and do measure toxicity when properly done and are an invaluable aid to discerning if toxicity is causing impact. Toxicity tests should be used to measure toxic stress and field studies should be used to measure the response of the community to that stress. If microcosms can accurately measure community interactions resulting from changed species composition, then they are useful tools for measuring toxicity. If not, then the effort is better spent doing single species tests and field studies. Just the mere presence of some species in a test vessel does not make the test more valid - it may make it less valid.

We are frequently reminded of the limitation of single species toxicity tests. We must be equally cautious of the limitations of community measurements and microcosm tests.

D. I. Mount

RESUME

Les études en laboratoires sont faites pour prédire plusieurs types de réactions ayant lieu sur le terrain. Lorsque l'usage se rapporte à ce qui concerne la toxicité, des tests de toxicité sont souvent utilisés. Les tests de toxicité mesure la sensibilité des espèces et rien d'autre. Par conséquent, pour prédire les effets sur les communautés à partir de tels tests, seulement les effets qui sont directement causés par la sensibilité aux substances chimiques sont valides aux fins de comparaisons. La toxicité stresse les espèces les plus sensibles. La toxicité n'a pas un effet "non-stressant" sur les espèces les plus tolérantes. Cependant ces espèces les plus tolérantes peuvent changer en nombre, un résultat du décroissement des espèces les plus sensibles. De tels effets ne peuvent être prédits avec les tests de toxicité.

Les études sur le terrain ne mesurent pas seulement les changements du stress ayant trait à la toxicité mais aussi les changements dus à toutes les causes. Rarement les mesures sur le terrain peuvent prouver les causes des changements. Les tests de toxicité peuvent faire et font le mesurage de la toxicité lorsque proprement exécutés et sont d'un aide inestimable pour discerner si la toxicité a un impact. Les tests de toxicité doivent être utilisés pour mesurer le stress toxique et les études sur le terrain devraient être utilisés pour mesurer la réponse des communautés à ce stress. Si les microcosmes peuvent mesurer précisément les interactions des communautés résultant dans le changement de la composition des espèces, alors ils sont des outils utiles pour mesurer la toxicité. Sinon, l'effort fait est alors mieux utilisée en faisant des tests sur une espèce et des tests sur le terrain. Juste la simple présence de certaines espèces dans un "test-tube" ne rend pas le test plus valide- cela le rend peut-être moins valide.

Nous nous faisons fréquemment rappeler les limites des tests de toxicité d'une simple espèce. Mais nous devons être également prudents à l'égard des limitations des mesurages des communautés et des tests microcosmes.

A TOXICITY ASSAY SYSTEM BASED ON BEHAVIOURAL RESPONSES OF FISH TO NATURAL CHEMICAL STIMULI

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Behavioural assays are sensitive to sublethal levels of pollution but they usually require highly trained personnel and long observation periods. We describe a system that combines the sensitivity of a behavioural assay with commercially available automated monitoring equipment. The assay is based on the response of fish to natural chemical stimuli such as food odours or pheromones. These behavioural responses can be altered by toxic effects on the chemoreceptors, on the central nervous system, or on general health and thus monitor several aspects of the well being of the fish. The observation system consists of a lucite aquarium coupled to a recirculating water system, and an Opto-Varimex-Aqua activity tracking meter (Columbus Instruments, Columbus, Ohio 43204) interfaced to a microcomputer. When stimulus solutions are injected into the water circulation the response of the fish is monitored by the computer system, which is capable of discriminating and quantifying changes in seven behavioural parameters. Normal responses to stimuli are compared with the response of fish that have been exposed to pollutants. Long-term (up to 70 hr), automated monitoring of the behaviour of control or experimental animals can be carried out with a single initial program command and up to eight activity

tracking meters can be run by the computer. We have successfully used this technique to examine effects of reduced pH on the response of fathead minnows to chemical feeding stimuli. The potential exists for this bioassay system to be extensively used to test for effects of pollutants on the behavioural response of fishes to a number of important natural stimuli such as food odour, sex pheromones, and alarm substances from damaged skin. The system should be easily adapted to any toxicology laboratory, and will identify effects of pollution that have thusfar been difficult or impossible to assess.

Un système de test sur la toxicité
basé sur les réponses comportementales
des poissons face à un stimulus chimique naturel.
A. Dennis Lemly and R.J.F. Smith

RESUME

Les essais comportementaux sont sensibles à un niveau pré-mortel de pollution mais requièrent généralement un personnel hautement entraîné et de longues périodes d'observation. Nous décrivons un système qui combine la sensibilité du test sur le comportement avec l'équipement d'observation automatisé commercialement disponible. Le test est basé sur les réponses du poisson à un stimulus chimique naturel comme l'odeur de la nourriture ou de phéromones. Ces réponses comportementales peuvent être altérées par des effets toxiques sur les récepteurs chimiques sur le système nerveux central, ou sur la santé générale et ainsi on peut enregistrer différents aspects de la bonne forme des poissons. Le système d'observation consiste en un aquarium acrylique associé avec un système de recirculation de l'eau et d'un compteur enregistreur d'activités Opto-Varimex-Aqua (Columbus instruments, Columbus, Ohio 43204) en interface avec un micro-ordinateur. Lorsque la solution représentant le stimulus est incorporée à l'intérieur de l'eau en circulation, les réponses des poissons sont enregistrées par le système d'ordinateur qui est capable de discerner et de quantifier les changements de sept paramètres comportementaux. Les réponses normales aux stimuli sont comparées avec les réponses des poissons qui ont été exposés aux polluants. Durant une longue période (jusqu'à 70 heures),

l'enregistrement automatise des comportements du groupe contrôle ou d'animaux expérimentaux peut être exécuté avec la commande d'un simple programme initial et jusqu'à huit activités peuvent être enregistrées sur compteur dirigé par ordinateur. Nous avons avec succès utilisé cette technique pour examiner les effets d'une diminution du pH sur les réponses des vairons à grosse tête lors d'un stimulus chimique dans l'alimentation. Le potentiel existant pour ce système de tests biologiques peut-être largement utilisé pour tester les effets des polluants sur les réponses comportementales des poissons de même qu'un nombre important de stimuli naturels comme l'odeur de la nourriture, les phéromones sexuels et les substances d'alarmes émises lors de l'endommagement de la peau.

Le système pourra être facilement adapté pour n'importe lequel des laboratoires de toxicologie et identifiera les effets des polluants qui ont été jusqu'à maintenant difficiles ou presque impossibles à évaluer.

Simon C. J. Pedder and Edward J. Maly

Abstract

Present research involves the use of a video monitoring system which records the attraction-avoidance responses of juvenile brook trout, Salvelinus fontinalis to varying levels of acidity (H_2SO_4). Using a two chamber artificial stream apparatus, groups of twenty individuals are given the choice of treated or untreated waters for 96 hour intervals. The video monitoring system allows us to record the time spent in each area while recording effects on social behaviour, such as territoriality. An ultra sensitive lens permits observations during the darkness cycle of the photoperiod. This system will hopefully lead to a better understanding of the effects of various toxicants upon behaviour especially during long term exposure and acclimation to darkness.

Note: Results of undergoing research with acid solutions (H_2SO_4) will be reported at the workshop.

Simon C.J. Pedder and Edward J. Maly

RESUME

La présente recherche implique l'usage d'un système de contrôle vidéo qui enregistre les réponses d'attraction/repulsion chez la jeune truite de ruisseau, *Salvelinus fontinalis*, à une variation du niveau d'acidité (H_2SO_4). Utilisant un appareillage doté de 2 chambres et produisant un courant artificiel, le choix d'eau traitée et non-traitée est donné à des groupes de 20 individus durant une période de 96 heures. Le système de contrôle vidéo nous permet d'enregistrer le temps passé dans chaque section tout en enregistrant les effets sur le comportement social, comme la territorialité. Une lentille ultra-sensible permet d'effectuer les observations sur le cycle de la nuit de la photopériode. Ce système va, espérons-le, mener à une meilleure compréhension des effets de plusieurs toxiques sur le comportement, spécialement pour une exposition longue-durée et de l'acclimatation à la noirceur.

Note. Les résultats de la présente recherche avec les solutions d'acide (H_2SO_4) vont être présentés à l'atelier.

Laboratory Avoidance Assessments in Fish:
Greater Ecological Relevance and a New Computerized Tracking System

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Classical laboratory tests give estimates of pollution preference/avoidance in a homogeneous or "open-field" environment, but can have limited ecological relevance for many species. This methodology tells little of how pollutant preference/avoidance interacts with other factors (eg. shelter, food, physicochemical conditions, migration) that are present in nature. This research attempts to incorporate some of these interactions in a fairly simple laboratory test to give a better indication of how an animal's site preference could mediate behavioural responses to pollutants.

Individual fathead minnows (Pimephales promelas), especially males in breeding condition, tend to remain under a shelter. Tests with zinc were performed in sharp-gradient laminar flow tanks, both with and without a shelter present in the dosed side of the tank. Data were acquired by a newly developed computerized tracking system. Using a video camera linked to an IBM Personal Computer through a commercially available digitizing board, the system stores data in real-time on the location and distance travelled of a subject in an experimental arena. The software could be useful in a variety of experimental situations, but was primarily designed for the study of pollution avoidance in fish.

Fairly sophisticated statistical analysis of various locomotory parameters is possible. Preliminary analysis indicates that the presence of a shelter can delay or even negate an avoidance response to zinc in the open-field situation.

R. Korver, J. Sprague, and D. Noakes

RESUME

Les tests classiques de laboratoires donnent des estimés sur la préférence et l'évitement en regard de la pollution dans un environnement ouvert homogène, mais ils peuvent avoir une pertinence écologique limitée pour plusieurs espèces. Cette méthodologie en dit peu sur comment la préférence/évitement des polluants "inter-agissent" avec d'autres facteurs (i.e. les abris, la nourriture, les conditions physicochimique, la migration) qui sont présents dans la nature. Cette recherche essaie d'incorporer certaines de ces interactions dans un test de laboratoire raisonnablement facile pour donner une meilleure indication sur comment la préférence d'un animal pour un emplacement peut affecter les réponses de comportement face aux polluants.

Les individus de l'espèce des vairons à grosses têtes (*Pimephales promelas*), spécialement les males en état de reproduction, ont tendance à demeurer sous un abri. Des tests avec du zinc ont été effectués dans des réservoirs à gradient prononcé d'écoulement laminaire, les deux avec et sans la présence d'un abri du côté dosé du réservoir. Les données ont été obtenues par un système de tracé informatisé nouvellement développé. Utilisant une caméra vidéo reliée à un ordinateur personnel (IBM) au travers d'un tableau numérique disponible commercialement, le système emmagasine en temps réel les données de localisation et de la distance voyagée par un sujet dans un secteur expérimental. Le "software" peut être utile pour plusieurs situations expérimentales, mais a été originellement désigné pour l'étude de l'évitement de la pollution chez les poissons.

L'analyse de statistiques suffisamment sophistiquées de plusieurs paramètres de locomotion est rendu possible. Les analyses préliminaires indiquent que la présence d'un abri peut provoquer un délai ou même enrayer les réponses d'évitement face au zinc dans une situation homogène.

Ultra-clean techniques in assessing the effects
of metals on phytoplankton

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The necessity of using ultra-clean, metal-free techniques in measuring primary productivity in water from Batchewana Bay, Lake Superior and Little Turkey Lake, Ontario was evaluated. There was no significant difference in productivity in lakewater collected with a Van Dorn bottle (which has metal components) and with a metal free rubber-and-plastic hand pump; however, incubation in polycarbonate flasks gave much higher values than in glass flasks. The decrease in values in glass flasks may have been due to the leaching of metals from the glass flasks: although there was no detectable difference in the quantity of Cu, Pb or Ni leached from the two types of flasks the values were below the detection limit of our method. The addition of a mixture of metals (Cu (5 µg/L), Zn (30 µg/L), Ni (25 µg/L), and Pb (25 µg/L)) resulted in a more drastic decrease in productivity in polycarbonate than in glass flasks.

Techniques ultra-propres pour l'évaluation
des effets des métaux sur le phytoplancton.

P.T.S. Wong, H. Shear, Y.K. Chau, C. Nalewajko and S. Rhamey

RESUME

La nécessité d'utiliser des techniques ultra-propres, sans métaux, dans le mesurage de la productivité primaire de l'eau de la baie de Batchewana dans le lac Supérieur et du lac "Little Turkey" en Ontario, a été évaluée. Il n'y avait pas de différence significative de la productivité entre l'eau des lacs collectée à l'aide de bouteilles Van Dorn (qui contiennent des composantes métalliques) et celle avec une pompe manuelle en plastic/caoutchouc, sans métal; toutefois, l'incubation dans les fioles en polycarbonate donne des valeurs beaucoup plus élevées que dans les fioles en verre. Les baisses de valeurs dans les fioles en verre ont pu être dues à la libération de métaux par les fioles de verre: bien qu'il n'y est pas eu de différence détectable dans la quantité de Cu, Pb, Ni libérée par les 2 types de fioles, les valeurs étaient sous la limite détectable par notre méthode. L'addition d'un mélange de métaux (Cu (5 µg/l), Zn (30 µg/l), Ni (25 µg/l), et Pb (25 µg/l)) a résulté dans une baisse plus sévère de la productivité dans les fioles en polycarbonate que dans celles en verre.

Introduction

Phytoplankton are known to be sensitive to low concentrations of metal contaminants in the waters (Wong *et al.*, 1982; Rai *et al.*, 1981). For example, Fitzwater *et al.* (1982) have shown that small amounts of metal contaminants introduced during sampling and handling procedures could adversely affect ¹⁴C-primary productivity measurements in the open-ocean waters. Since sampling and incubation techniques in the Great Lakes rely on standard methods similar to those in oceans, the possibility of metal contamination is equally great, particularly in the oligotrophic Upper Great Lakes, where the trace metal and nutrient concentrations are extremely low. Our objective was to determine if ultra-clean sampling and experimental procedures are necessary in Lake Superior and in Little Turkey Lake.

Materials and Methods

Sampling sites -

We chose Batchewana Bay (latitude 46° 49'N; longitude 84° 39'W) in Lake Superior and Little Turkey Lake (latitude 46° 45'N; longitude 84° 30'W) in Ontario for our July-August, 1983 studies because both are oligotrophic and accessible (Vollenweider *et al.*, 1974).

Sampling water -

We examined several types of water samplers for possible contamination from their metal components. The most commonly used Van Dorn bottle (General Oceanics) has a number of metal parts and fittings which would be

difficult to remove or replace with non-metal parts. The Niskin bottle (GM Manufacturing) has fewer metal parts (2 pins, 1 spring and 1 clamp) but is available only in a 2.5 L size. The Go-Flo ball-valve bottle (General Oceanics) was difficult to operate manually especially in a small boat. The "March" pump (March Manufacturing Inc.) could be used to pump a large volume of water at a specific depth. The pump has no metal components. Unfortunately a generator is required for pumping. The rubber-and-plastic hand-operated pump (ITT Industries) has no metal parts, is light, and can be operated manually. After experimenting with various bottles in the field, we decided to use the Van Dorn bottle as the conventional method of taking water samples, and the rubber-and-plastic hand pump as the metal-free method of taking water samples.

Water was collected at a depth of 4.9 m, mixed in a plastic carboy and dispensed into 500 mL erlenmeyer glass or polycarbonate flasks. Four flasks were housed in one flask holder and lowered into 4.9 m depth for in-situ incubation (Figure 1) with $^{14}\text{C-HO}_3$.

Optical properties of flasks -

Light penetration into the flasks was investigated in the laboratory. Flasks were illuminated with quartz-halogen lamps (Philips 12013R) and light intensities inside the flask were measured with a Biospherical Instruments Model QSL-100 quantum meter. Spectral composition was investigated with an ISCO model SR spectroradiometer.

Mooring -

Three types of moorings were compared for ease in handling and

retrieval of the samplers (Figure 1). The spar buoy had a radar reflector attached to a plastic buoy on a 6 foot spar. The sampler was stabilized with a half concrete block. We found that it was difficult to retrieve the sampler because of the weight of the spar buoy itself, and the necessity to detach and reattach the spar buoy to the anchor line in order to retrieve the sampler. This was found to be very awkward for two people in a small boat in rough weather. In addition, the radar reflector was not essential to our work. The tether buoy had a similar problem in that retrieval of the samples required detaching and reattaching the anchor rope each time. The dumbbell buoy was convenient for retrieving the sampler simply by undoing the knot from one end of the pipe. The concrete block was subsequently replaced with a smaller, plastic-coated lead weight. The dumbbell buoy was used in all our experiments.

Primary productivity -

To reduce metal contamination from ^{14}C -bicarbonate, we used high activity stock (56 mC/m mole and 5 mCi/mL) from Amersham Co. This was diluted to a working solution of 50 $\mu\text{C/mL}$ with double distilled water and stored in acid-cleaned Teflon bottles. For experiments, we added 10 μC to a 500 mL sample. The flasks were tightly capped with rubber stoppers which were wrapped with teflon tape. After 4-hour incubation, 500 mL sample was filtered on a 0.45 μ cellulose acetate filter, and rinsed rapidly with 100 mL of double distilled water. Filters were dissolved in 15 mL PCS scintillation counting fluor (Amersham/Searle) and counted in a liquid scintillation counter (Beckman model LS8100).

Cleaning -

All bottles, flasks, graduated cylinders, pipettes and tubings were sequentially soaked overnight in Decon (BDH Ltd.), dilute nitric acid (4-10%), 0.02 M EDTA, and rinsed several times with double distilled water.

Metal analyses -

Cu, Pb, Ni and Zn were solvent-extracted and analyzed in a graphite furnace, atomic absorption spectrophotometer with a detection limit of 1 $\mu\text{g/L}$ (Environment Canada, 1979).

Results and Discussion

Fitzwater *et al.* (1982) identified several potential sources for metal contamination that could adversely affect the productivity measurement in the waters. These included water samplers, incubation flasks, and radioactive isotopes. To test the effects of possible metal contamination from the sampling bottles, we compared the primary productivity in water from Batchewana Bay taken with Van Dorn bottle or rubber-and-plastic hand pump and incubated with ^{14}C in polycarbonate flasks. The results (Table 1) indicate that the productivity in water taken with the "metal-free" technique was slightly higher than in water taken with the conventional Van Dorn bottle but the results are not statistically different as analyzed by the Student-Newman-Keuls test at 95% confidence limits. Similarly, there was no significant difference in productivity in water from Little Turkey Lake using two water sampling techniques (Table 2). Hence, the leaching of

metals, if any, from the Van Dorn bottle did not inhibit the phytoplankton significantly.

In the course of the experiments, we realized that the dispensing of water into the incubation flasks on board a diesel fuel boat could lead to the contamination of water by metals and organic compounds from the vapor of the diesel fuel. To avoid this atmospheric contamination, we devised an in-situ incubation technique. We connected the 4 incubation flasks with 2 glass U-tubes (Figure 2), stoppered tightly and lowered the samplers to 4.9 m depth. A porcelain messenger was sent down to break the U-tubes and allowed the water to enter the flasks by hydrostatic pressure. This technique avoided metal contamination from the water-sampling bottles and from the atmosphere.

Metal contamination could also come from the incubation flasks. Fitzwater *et al.*, (1982) reported that even after vigorous cleaning, significant amounts of Cu leached from the glass flasks and significantly depressed primary productivity. To verify this observation, we compared the productivity in water incubated in glass or polycarbonate flasks using the above in-situ technique. The productivity in waters from Lake Superior and Little Turkey Lake incubated in polycarbonate flasks was indeed much higher than in glass flasks (Table 3). A similar observation was reported by Carpenter and Lively (1980) and was attributed to the relatively low metal leaching from the polycarbonate flasks.

Experiments were carried out to determine whether the low productivity

in glass flasks was due to the leaching of toxic metals from the walls of glass flasks. Water samples from Batchewana Bay were collected either with Van Dorn bottle or rubber-and-plastic hand pump and incubated in polycarbonate and glass flasks for 4 hours. The concentrations of Cu, Pb, Ni and Zn in the waters were determined. Results (Table 4) indicate that Cu, Pb and Ni levels were below detection limits of 1 $\mu\text{g/L}$ in all the samples. There was no significant difference in the Zn concentrations of 6.0 - 7.5 $\mu\text{g/L}$. Similarly, water samples from Little Turkey Lake contained <1 $\mu\text{g/L}$ of Cu, Pb and Ni and 16.0-17.5 $\mu\text{g/L}$ of Zn (Table 5). Extending the incubation time from 4 hours to 28 days did not increase the concentration of the metals.

To test the possibility that the observed increase in the productivity in polycarbonate flasks was caused by more effective light penetration into the polycarbonate bottles (Ilmavirta and Hakata, 1972), we compared both the light intensity and spectral composition inside the two types of flasks (Table 7). The polycarbonate flasks proved to be less transparent than glass, and showed a trend of increasing absorbence with increasing wavelenth in the 525-675 nm range. Overall, photosynthetically available energy (400-700 nm) inside a polycarbonate flask would be 84.9% of that in a glass flask. Since incubations were carried out at a depth of 4.9 m, we were working in the region of light-limited photosynthesis. Therefore, the lower light intensity inside the polycarbonate flasks would decrease photosynthesis and not increase it, as observed here.

In conclusion, the only logical explanation of the significantly lower productivity in glass flasks must involve the assumption that metals, or, possibly, other substances, leach from glass and inhibit photosynthesis. Our analytical methods are not capable of detecting metals at $<1 \mu\text{g/L}$, but some metals, eg. Cu, can be toxic to phytoplankton at concentrations $<1 \mu\text{g/L}$ (Hodson et al., 1979 and Reuter et al., 1979). It is also possible that the synergistic effects of metals or of a metal not included in our analysis were responsible for the toxicity.

The emphasis by Fitzwater et al. (1982) on using the ultra-clean technique in measuring productivity in oligotrophic waters is thus valid. As shown in Table 6, addition of a metal mixture of Cu, Zn, Ni and Pb at the Great Lakes Water Quality Objectives levels (IJC, 1976) reduced the productivity by 39 to 74%. While the standard Van Dorn sampler proved not to be a source of contamination, incubation in glass bottles is not acceptable. The drastic decrease in productivity must be attributed to leaching of toxicants from the glass. However, the decreased metal toxicity in glass flasks could be explained by higher adsorption of the metals onto the flask walls (Robertson, 1968). It is evident that the use of glass containers is not appropriate for ^{14}C productivity experiments in oligotrophic lakes.

Acknowledgements

The authors would like to thank Water Quality Branch, Ontario Region for chemical analyses; Engineering and Technical Support Division for construction of the in-situ samplers and Ship Division for ship and technical support.

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Table 1. Effect of water sampling techniques on primary productivity in Lake Superior water

Water sampling techniques	No. of samples	Primary productivity dpm/500 mL (mean $\pm$ S.E.)
Conventional**	9	6,834* $\pm$ 302
Metal-free***	5	7,301* $\pm$ 614

* No significant difference in Student-Newman-Keuls test at 95% confidence limits

** Van Dorn bottle

*** Rubber-and-plastic hand-operated pump.

Table 2. Effect of water sampling techniques on primary productivity in Little Turkey Lake water

Water sampling techniques	No. of samples	Primary productivity dpm/500 mL (mean $\pm$ S.E.)
Conventional**	8	18,100* $\pm$ 3,526
Metal-free***	8	21,872* $\pm$ 828

* No significant difference in Student-Newman-Keuls test at 95% confidence limits

** Van Dorn bottle

*** Rubber-and-plastic hand-operated pump.

Table 3. Comparison of primary productivity in waters from
Lake Superior and Little Turkey Lake using in-situ technique
with glass and polycarbonate flasks

Location	No. of Samples	Primary productivity		P.C./glass
		(dpm/500 mL; mean $\pm$ S.E.)		
		glass flask	polycarbonate flask	
Lake Superior	10	4,125 $\pm$ 205	13,715 $\pm$ 1,076	3.3
Turkey Lake	13	42,403 $\pm$ 5,271	110,902 $\pm$ 20,839	2.6

Table 4. Effects of water collection methods and incubation
flasks on Cu, Pb, Ni and Zn concentrations ($\mu\text{g/L}$) in
water from Batchawana Bay

Water Collection Method	Type of flask	Sample Number	Cu	Pb	Ni	Zn ($\bar{x} \pm \text{S.D.}$)
Metal-free	polycarbonate	2	<1	<1	<1	$6.0 \pm 1.4^*$
Conventional	polycarbonate	2	<1	<1	<1	$7.5 \pm 0.7^*$
Metal-free	glass	2	<1	<1	<1	$7.5 \pm 2.1^*$
Conventional	glass	2	<1	<1	<1	$6.5 \pm 0.7^*$

*No significant differences in Student-Newman-Keuls test at 90% confidence limits

Table 5. Effects of water collection methods and incubation flasks
on Cu, Pb, Ni and Zn concentrations ($\mu\text{g/L}$)
in water from Little Turkey Lake

Water Collection Method	Type of flask	Sample Number	Cu	Pb	Ni	Zn ($\bar{x} \pm \text{S.D.}$)
Metal-free	polycarbonate	4	<1	<1	<1	$16.5 \pm 2.1^*$
Conventional	polycarbonate	4	<1	<1	<1	$16.0 \pm 0^*$
Metal-free	glass	4	<1	<1	<1	$17.5 \pm 0.6^*$
Conventional	glass	4	<1	<1	<1	$16.3 \pm 1.0^*$

*No significant differences in Student-Newman-Keuls test at 90% confidence limits

Table 6. Effects of a metal mixture on
primary productivity in waters from
Lake Superior and Little Turkey Lake

Water	Addition	Flask	Primary productivity (dpm/500 mL)	% of control
Lake Superior	None	Glass	4,125	100
"	metals*	"	2,425	59
"	none	Polycarbonate	13,715	100
"	metals	"	3,704	27
Turkey Lake	None	Glass	42,403	100
"	metals	"	25,935	61
"	none	Polycarbonate	110,902	100
"	metals	"	29,058	26

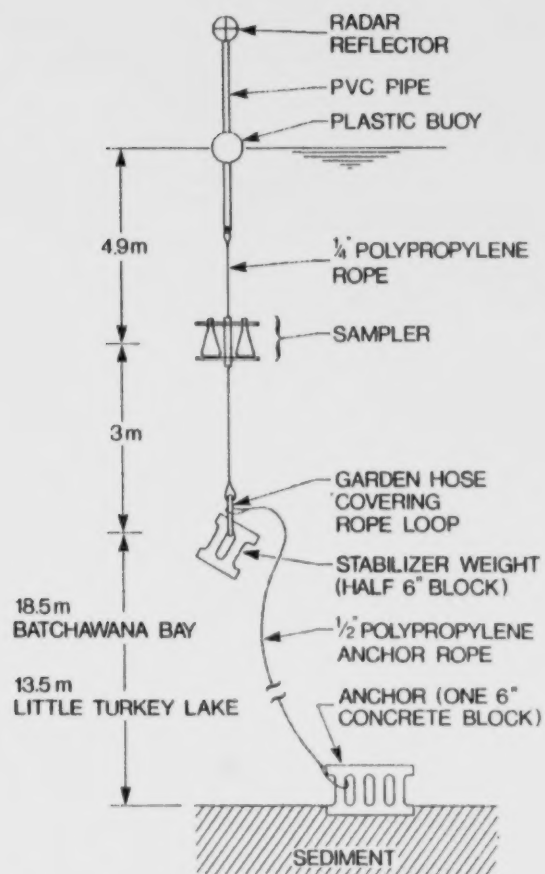
*Metal mixture contained Cu (5), Zn (30), Ni (25) and Pb (25) $\mu\text{g/L}$.

Table 7.

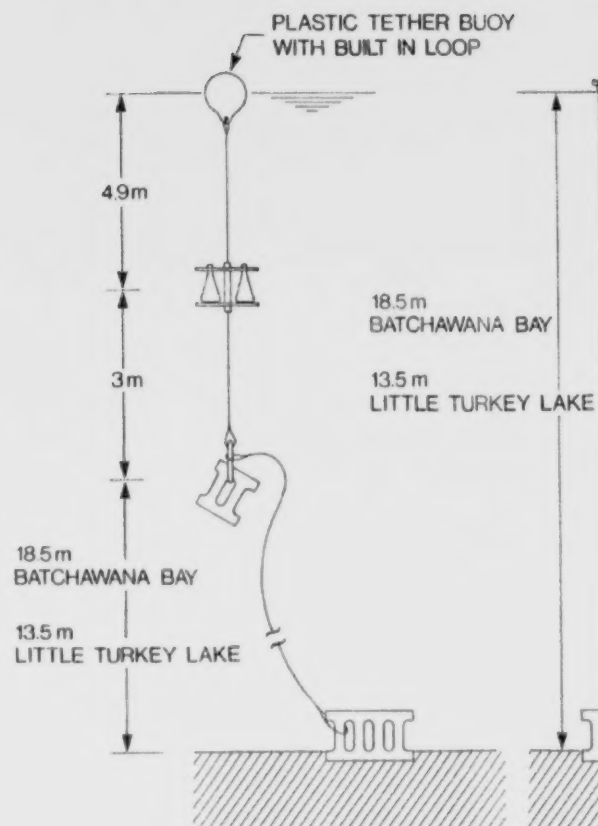
Light intensity and spectral distribution inside incubation flasks, illuminated with quartz halogen lamps. Measurements were made at a fixed point inside the flask and the probe was lowered at right angles to the light source.

Light energy, quanta $\text{cm}^{-2} \cdot \text{sec}^{-1}$	Polycarbonate flask	Glass flask
	0.747 ± 0.05	0.880 ± 0.04
Spectral composition		
$\mu\text{W} \cdot \text{cm}^{-2} \cdot \text{nm}^{-1}$		
425 nm	2.07	2.07
450	3.39	3.60
475	5.50	6.00
500	7.74	8.16
525	10.15	11.09
550	12.88	14.49
575	15.13	17.00
600	17.00	19.55
625	19.64	22.44
650	22.10	25.16
675	20.74	27.20
700	26.10	21.60
725	28.44	30.06
750	30.45	32.28

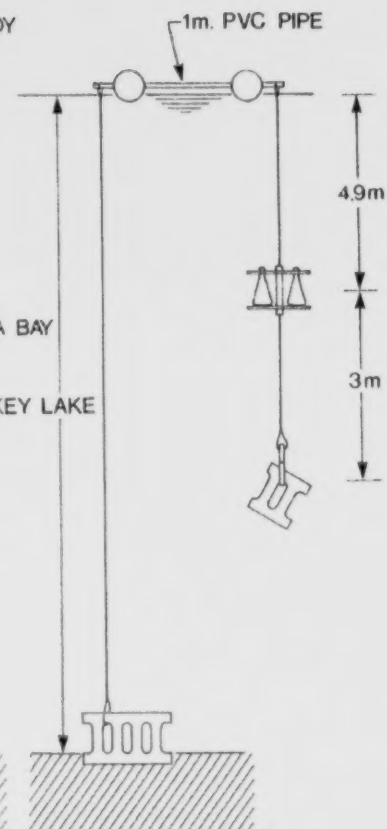
① SPAR BUOY



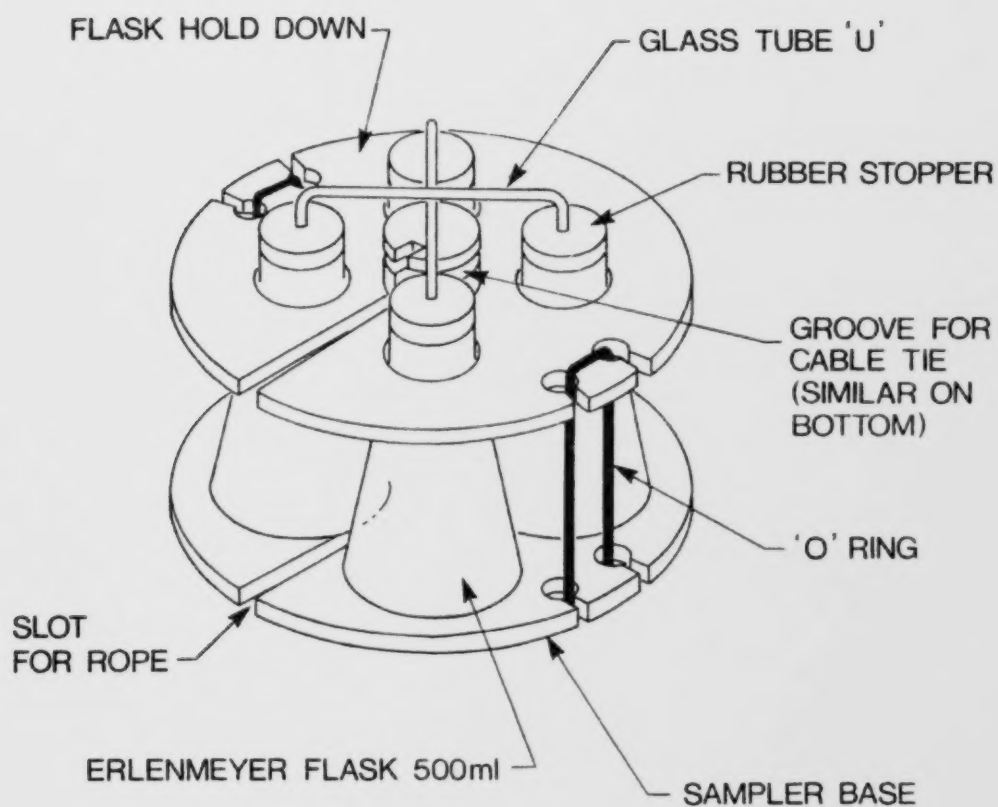
② TETHER BUOY



③ DUMBBELL BUOY



THREE TYPES OF MOORING



IN-SITU SAMPLER

ASSESSMENT OF RISK CRITERIA WITH LABORATORY AND FIELD DATA

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Pesticide registration in the United States is based on the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA) and promulgated through regulations developed by the Environmental Protection Agency. One part of the regulation requires an environmental risk assessment for aquatic life. The risk assessment is accomplished through a comparison of laboratory toxicity data to risk criteria.

The goal of this project was to go beyond the laboratory data-risk criteria comparison by associating data from laboratory toxicity tests, on-site bioassays and on-site invertebrate population monitoring obtained from a natural pond treated with the mosquito larvicide chlorpyrifos.

The study was conducted in 1983 in a pond located in northern Hennepin County near Minneapolis, Minnesota. Chlorpyrifos was applied by the regional mosquito control district. The insecticide concentration in the water was obtained by pre- and post-application measurements. Biological measurements included on-site bioassays of 6 resident invertebrate taxa and monitoring the populations of 40 taxa. The population monitoring began on 25 May and continued through 13 October 1983.

Nine risk criteria applicable to the FIFRA based freshwater risk assessment were analyzed. Of the nine, six were evaluated with data from this study. This evaluation showed that the criteria were predictive of the effects seen in the pond.

Richard L. Anderson, Ph.D.

RESUME

L'enregistrement des pesticides aux Etats-Unis est basé sur l'Acte Fédérale des Insecticides, Fongicides et Rodenticides (AFIFR) promulgué par l'entremise de règlements développés par l'Agence de Protection de l'Environnement. Une part des règlements requiert l'évaluation des risques pour l'environnement quant à la vie aquatique. L'évaluation des risques est exécutée par la comparaison des données de toxicité en laboratoire avec des critères de risques.

Le but de ce projet était d'aller au-delà de la comparaison des critères de risques en laboratoire en associant les données; des tests de toxicité de laboratoire, des tests biologiques sur le terrain et de l'enregistrement sur le terrain des populations d'invertébrés obtenues d'un étang naturel traité avec le larvicide pour moustiques "chlorpyrifos".

L'étude a été conduite en 1983 dans un étang situé au nord du comté d'Hennepin près de Minneapolis au Minnesota. Le "chlorpyrifos" a été appliqué par le district régional de contrôle des moustiques. La concentration en insecticide de l'eau a été obtenue par le mesurage avant et après l'application. Les mesurages biologiques incluaient les tests biologiques sur le terrain de 6 taxa d'invertébrés sédentaires et l'enregistrement des populations de 40 taxa. L'enregistrement des populations commençait le 25 mai et continuait jusqu'au 13 octobre 1983.

Neuf critères de risques applicables à l'évaluation des risques en eau douce basée sur l'AFIFR ont été analysés. Sur les neuf, six ont été évalués avec les données de cette étude. Cette évaluation démontrait que les critères constituaient une prédiction des effets vérifiés dans l'étang.

BIOTESTS AVAILABLE IN EPS ENVIRONMENT CANADA LABS,
THEIR LINK TO THE CONCEPT OF ECOTOXICOLOGY AND
COST EFFECTIVE PRACTICALITIES

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ABSTRACT

The impact of technological growth on the environment has reached the point where a fundamental rethinking of technical means used to assess toxicity has become necessary. Now more than ever ecotoxicology - an alliance of biology and chemistry - is proving an indispensable tool both for determining the effects of given insults to the environment and for identifying their causes. Coupled with its physical chemical expertise, EPS Environment Canada labs now have the basic biological tools to act in this new scientific endeavour. Cost-efficient practicalities however must be considered for each environmental prospect anticipated. This presentation through the use of simple schematic diagrams depicts ecotoxicity as a spiraling aggressor affecting the ecosystem and superimposes the 22 different types of biotools available in these canadian laboratories. Practical cost-efficient considerations are also presented with these overlays such as chrono-responses, effort in man days and wastewater volume required per biotest.

Les tests biologiques disponibles dans
 les laboratoires d'EPS Environnement Canada;
 leurs liens au concept de l'écotoxicologie et les
 coûts effectifs de la pratique.

N. Bermingham, C. Blaise, R. Van Coillie and R. Vezeau

RESUME

L'impacte de la croissance technologique sur l'environnement a atteint le point où, par nécessité, les moyens techniques utilisés pour évaluer la toxicité doivent être fondamentalement repensés. Maintenant plus que jamais l'écotoxicologie - une alliance de la biologie et de la chimie - se veut un outil indispensable aussi bien pour déterminer les effets certains des offenses sur l'environnement que pour en identifier les causes. En association avec ses compétences en physique et en chimie les laboratoires d'EPS Environment Canada ont maintenant les outils biologiques de base pour oeuvrer dans ce nouvel effort scientifique. Quoi qu'il en soit, l'efficacité en regards des coûts de la pratique doit être considérée pour chaque perspective environnementale anticipée. Cette présentation par l'usage de simple diagramme schématique décrit l'écotoxicologie comme un agresseur grandissant affectant l'écosystème et surtaxant les 22 différents types d'outils biologiques disponibles dans ces laboratoires canadiens. Les considérations quant à l'efficacité des coûts pratiques sont aussi présentées avec ces différentes parties comme les réponses dans le temps, l'effort journalier de l'homme et le volume d'eau résiduel requis pour chaque test biologique.

Evaluation of the Toxicity of Spruce Budworm Insecticides to Aquatic Invertebrates

David G. Poirier and G. A. Surgeoner*

Abstract

The toxicities of two registered and two candidate forest insecticides, to six families of aquatic invertebrates were evaluated using continuous flow laboratory and field bioassay systems. Permethrin (LC50s 0.99 to 9.24 ug/l) was discovered to be 10 to 250 times more toxic than fenitrothion (LC50s 42.38 to 283.70 ug/l) which in turn was 0 to 16 times more toxic than aminocarb (LC50s 344.43 to 1275.60 ug/l) and mexacarbate (LC50s 98.80 to 1503.65 ug/l). Results from lab and field assay systems did not differ significantly in slope of probit line or LC50 values in most cases. Results of experiments carried out during an experimental aerial spray of fenitrothion indicated moderate impact on aquatic invertebrates. Caged insect mortalities were accurately predicted by the lab and field bioassay results. These assay apparatus offer great ease and flexibility in evaluating relative toxicities of various chemicals, chemical formulations, and the different effects of environmental parameters (eg. temperature, pH) on aquatic organisms.

Les toxicités de deux insecticides enregistrés pour usage silvicultural et de deux insecticides candidats ont été évalués pour six espèces d'invertébrés aquatiques utilisant un système d'analyse de flux continu dans le laboratoire et un système d'analyse dans la forêt. Permethrin (CL50 0.99 à 9.24 ug/l) était significativement plus toxique (10 à 250 fois) que fenitrothion (CL50 42.38 à 283.699 ug/l) qui était encore 0 à 16 fois plus toxique qu'aminocarb (CL50 344.43 à 1275.60 ug/l) et mexacarbate (CL50 98.80 à 1503.65 ug/l). Les résultats des systèmes d'analyse dans le laboratoire et dans la forêt n'ont pas différés significativement dans les pentes des lignes de probits ou pour la plupart dans les CL50s. Les résultats des essais faits pendant une application aérienne expérimentale de fenitrothion ont indiqués des impacts modérés sur les invertébrés aquatiques et les mortalités des insectes cagés ont été prédits précisément par les résultats des analyses dans le laboratoire et dans la forêt. Ces appareils d'analyse offrent beaucoup de flexibilité dans l'évaluation des toxicités relatifs de divers produit chimique, des formulations chimiques et des effets de divers paramètres, comme temprature et pH, sur les organismes aquatiques.

Introduction

Aquatic biologists have expressed concern over the potential detrimental effects of forest insecticides on aquatic ecosystems. With the continued high infestations of spruce budworm in North America, the aerial applications of these chemicals will continue, due to the absence of other effective control measures. Modifications to the aerial spray programs continue, however, a suitable bioassay apparatus which can accurately predict impacts of candidate or registered insecticides on stream ecosystems has not been developed.

This summary outlines a series of bioassay experiments which simulate the environmental factors encountered during aerial applications of spruce budworm insecticides. These results will be compared to caged insect, drift and bioassay studies carried out during an experimental aerial spray of fenitrothion. The relative toxicities of two registered and two candidate spruce budworm insecticides will be evaluated using the laboratory and field bioassay units and the potential uses of these bioassay procedures will be indicated.

Materials and Methods

The laboratory and field bioassays were designed to simulate the environmental factors encountered during spruce budworm control programs. Lab bioassays were conducted in environmental chambers with a light regime of 16h light and 8h darkness; the temperature regime was set at 16°C (day) and 8°C (night). The insecticides were used in the spray formulations (Table 1) rather than as the technical material. Test organisms were aquatic invertebrates selected to represent a wide range of taxa and trophic niches (Table 2). The invertebrates were exposed to the toxicants for only

1h, and observed for mortality for 48h. This exposure simulated the "pulse" dosage noted while monitoring actual aerial spray programs (Coady, 1978; Holmes, 1979; Kingsbury, 1981).

The bioassay apparatus used in the lab was designed by Rodrigues and Kaushik (1984). It consisted of a 10 l aquarium, an aquarium power filter, a glass mixing chamber, and a glass trough all supported on a wooden frame. This apparatus had a flow through mode in which diversion of contaminated water could occur while dosing, and a recirculation mode during the remainder of the 48h observation period. The recirculation mode reduced the amount of water required for experimentation while maintaining a flowing water column for the test organisms.

The field apparatus was modified from that of Travis and Schuchman (1968). It consisted of a parallel series of vinyl eaves troughs through which water was diverted from a stream (Icewater Creek, Ontario) (Figure 1a). The water was diverted to head tanks using a gravity feed system and then supplied to each trough at a rate of 116 l/h. The dosing apparatus consisted of 4 l mariotte bottles calibrated to drain in 1h (Figure 1b). The invertebrates were placed in the troughs and exposed to the insecticide as previously indicated.

The experimental aerial spray at Icewater Creek was conducted by the Forest Pest Management Institute, in Sault Ste Marie, Ontario. Fenitrothion was applied by Ag-Truck airplane at a rate of 280g A.I./Ha with no setback distance from the creek. This procedure was designed to produce a "worst case" scenario in order to evaluate all impacts. Three experiments were used to evaluate the impact on non-target aquatic invertebrates. Aquatic invertebrates (Table 2) were placed in small cages (15cm x 3cm x 3cm) in the stream 24h prior to the spray and were removed 24h after the spray. The number of dead invertebrates were recorded at this time. Samples of drifting insects

and stream water were taken at 15 minute intervals for the duration of the spray program. Drift samples were examined for the number of aquatic larvae present and stream water was analysed for the concentration of fenitrothion present. Two hundred litres of contaminated stream water were sampled between 0 and 30 minutes post-spray and were used to conduct a standard 24h laboratory bioassay (24h dose period) with the same insects used in the caged insect studies.

Analysis of data included testing probit lines for parallelism (Draper and Smith, 1966) and testing 48h LC_{50} values for significant differences (Sprague and Fogels, 1976). This latter method was examined and approved by J.J. Hubert of the Department of Statistics at the University of Guelph.

Results and Discussion

For the purpose of this summary all the LC_{50} s and slopes of the probit lines are presented in tabular form (Table 3; Table 4), but only a portion of the data will be discussed to indicate a few general trends. Two major observations had a significant bearing on these results, and indeed on the authors decision to utilize these assay procedures. The first observation was that there was not a single control mortality over the two years of study in the laboratory or the field. This indicated that the trough systems were readily acceptable to the invertebrates and placed no undue stress on them. The second result was the initiation of a drift response with the organisms in the troughs. This behavior has been noted in the field in response to the introduction of toxicants (Holmes, 1979; Kingsbury, 1981), and indicates that the test species were responding naturally in the troughs. This drift response was initiated at insecticide concentrations of 0.5 ug/l permethrin and 10 ug/l for the other three

chemicals examined.

A trend easily recognized in the data involves the relative toxicities of the four forest insecticides. In all cases permethrin was significantly more toxic to aquatic invertebrates than the other chemicals (Figure 2).

This trend was noted in the lab and field systems with the results being:

permethrin > 10 to 250 x fenitrothion
 fenitrothion > 0 to 10 x mexacarbate
 mexacarbate $\geq$ 0 to 4 x aminocarb.

In only a few cases were there any significant differences in the slope of the probit lines between chemicals for the same test organism. These differences occurred primarily with permethrin, and may have been due to the different mode of action and the physical properties of the pyrethroids. It is widely believed that the pyrethroids act along the nerve axon, in contrast with the carbamates and organophosphorus insecticides which act to inhibit acetylcholinesterase in the nerve synapse. The result may be a faster response in the test organism and thus a steeper slope. In the field system, the pyrethroids tendency to adsorb onto any surface (Sharom and Solomon, 1981)(in this case the vinyl troughs) may have caused the differences in slope (Table 4; Figure 2) (pers. comm. J.B. Sprague 1985). In all cases of differential slopes a few more repetitions would probably reduce the variance of the slopes and improve the confidence intervals on the LC_{50} values. Due to time limitations on this study only one or two repetitions could be carried out for each combination of insecticide and test species.

The invertebrate groups showed different sensitivities to the various insecticides examined. The stoneflies were most sensitive to fenitrothion, permethrin and mexacarbate, however, the mayflies were equally as sensitive to fenitrothion and mexacarbate and more sensitive to aminocarb. In general the use of mayflies and stoneflies as indicator species would give a fairly sensitive assay of aquatic impacts. They have been shown to be most

sensitive to toxicants in other studies (Varty, 1978; Kingsbury and Kreutzweiser, 1979), and they were the most sensitive species in this study.

The experiments carried out during the aerial spray program served two purposes. Firstly, they demonstrated what actually occurs in a stream during an aerial spray program (eg. "pulse" exposure to toxicant, invertebrate drift, etc.), and secondly, they generate results to which the lab and field bioassay results could be compared. Fenitrothion concentration was plotted against invertebrate drift (Figure 3) to exhibit the interactions of these two responses in the natural environment. Fenitrothion concentration peaked rapidly at 31 ug/l and fell to 3.4 ug/l within 1h after the spray. This concentration fell further to 0.5 ug/l after 6h. Invertebrate drift increased rapidly peaking approximately 3h after the spray with numbers falling to prespray levels within 24h. This 1h peak exposure period has also been observed in previous studies (Holmes, 1979; Varty, 1978). These data supported the methodology chosen, rather than a standard 48h dosing period. One of the major objectives was the accurate portrayal of the conditions encountered during an aerial application of chemical insecticide. Dosing the organisms with contaminated stream water (fenitrothion concentration = 21.0 ug/l) for 24h caused twice the mortality encountered with invertebrates caged in the stream (Table 5). There is a major difference in results obtained from the two different assay techniques. Laboratory and field bioassays conducted using the 1h dosage generated results which were not statistically different from each other, and which accurately predicted the mortality which occurred with insects caged in the stream during the spray (Figure 4). The major argument put forth in this case is that insects released into the drift are carried along within the pulse of insecticide and thus are exposed to the higher concentrations for a longer period of

time. In this study the increase in drift lagged 1 - 2 hours after the major peak of insecticide had passed and were drifting in a concentration of fenitrothion of 3.8 ug/l, rather than the 31.0 ug/l observed at peak exposure time. This type of delayed response was also observed with aerial applications of Matacil (Holmes, 1979), but with the rapid knock-down properties of the pyrethroids the invertebrates may indeed be drifting within the peak concentration of permethrin after an aerial application (Kingsbury and Kreutzweiser, 1980).

In conclusion there are 4 major points to be made. Firstly we observed that lab bioassay and controlled field assay results can accurately predict impacts occurring in natural aquatic ecosystems after aerial application of insecticides. Secondly, there can be major impacts on aquatic invertebrate populations when using fenitrothion or permethrin for budworm control. We observed up to 25% mortality of some species with fenitrothion, and one can expect up to 75% mortality of sensitive species with permethrin. Matacil and zectran offer a 2 to 10 fold safety margin; therefore, aquatic impacts caused by these two chemicals would be expected to be minimal. The third major point involves increasing the efficacy of budworm control while reducing impacts on aquatic ecosystems. Since there are such wide safety margins with the two carbamate insecticides, it is feasible to reduce the 1 km buffer zones (presently required by law) along streams, and in such a way reduce the reservoir of budworm in these areas and still maintain minimal aquatic impacts. Finally, these assay systems offer great flexibility for evaluating relative toxicities of different chemicals to aquatic organisms. A brief experiment to evaluate the different toxicities of three formulations of permethrin (EC, ULV, and microencapsulated) to dragonfly naiads indicated that the microencapsulated formulation was 5 times less toxic than the others. This experiment took only 48h to finish, and

included a repetition of the results. This flexibility can be easily used to evaluate the effects of different environmental parameters on chemical toxicities, and thus more accurately predict what occurs in other situations.

Acknowledgements

I would like to thank the researchers at the Forest Pest Management Institute, and especially Mr. P.D. Kingsbury for their assistance during this project. Special thanks goes to Craig Logan for his patience and help over the past two summers, and to Dr. G.A. Surgeoner for his guidance. This study was made possible through Environment Canada grant number 854-26.

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Figure 1a: Continuous flow bioassay apparatus used in the field toxicity studies.

- 1 - 5 cm diameter water supply pipe
- 2 - 200 L head tank
- 3 - vinyl holding troughs for test organisms
- 4 - drainage troughs
- 5 - support stand

Figure 1b: Dosing apparatus and water valves used to control and distribute toxicant during the bioassay experiments.

- 1 - 4 L Marriotte bottle
- 2 - water control valves
- 3 - mixing funnels
- 4 - vinyl holding troughs for test organisms
- 5 - support stand

Figure 2a: Percent mortality curves for the toxicities of four forestry insecticides to aquatic invertebrates in the laboratory.

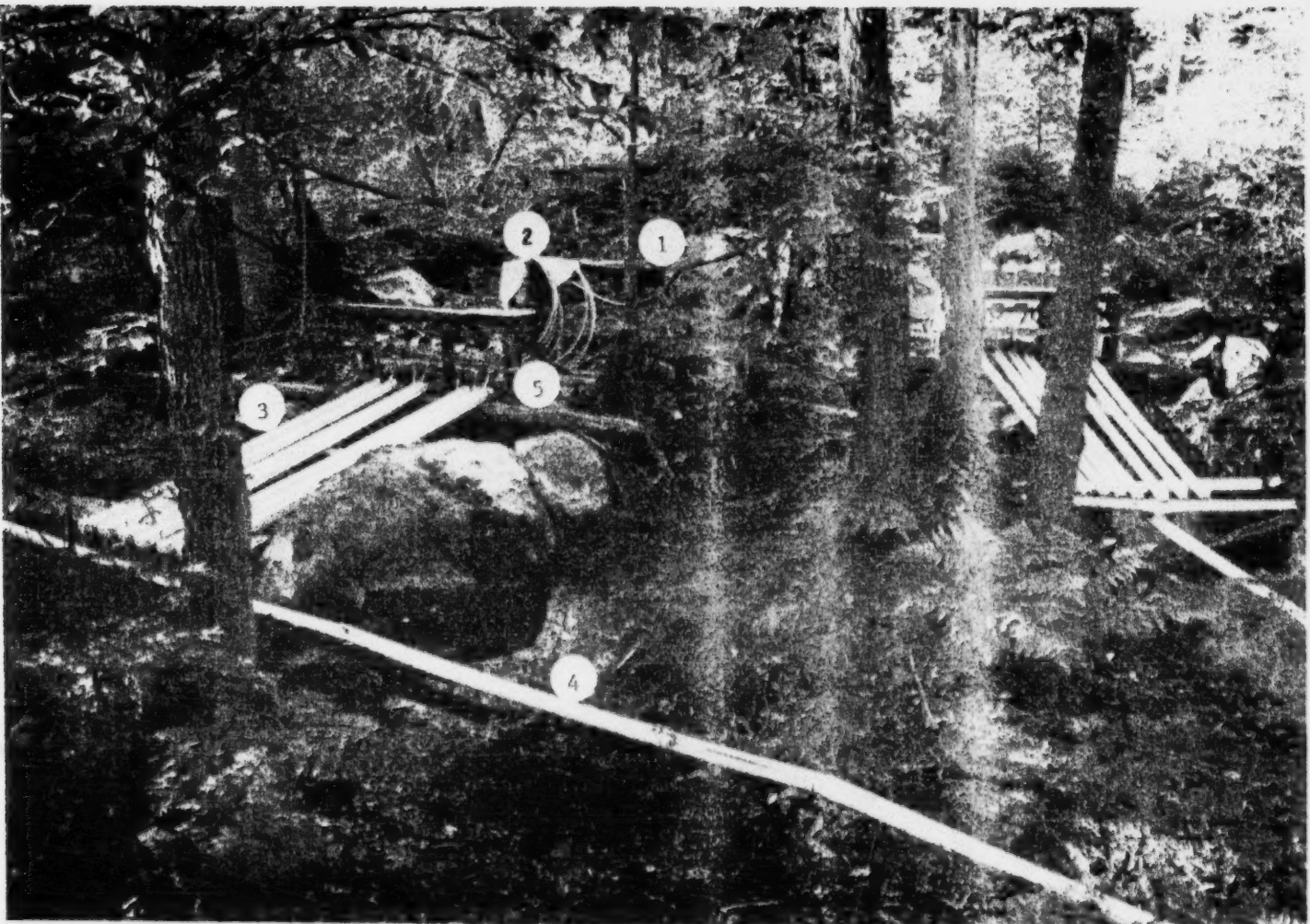
Figure 2b: Percent mortality curves for the toxicity of four forestry insecticides to aquatic invertebrates in the field.

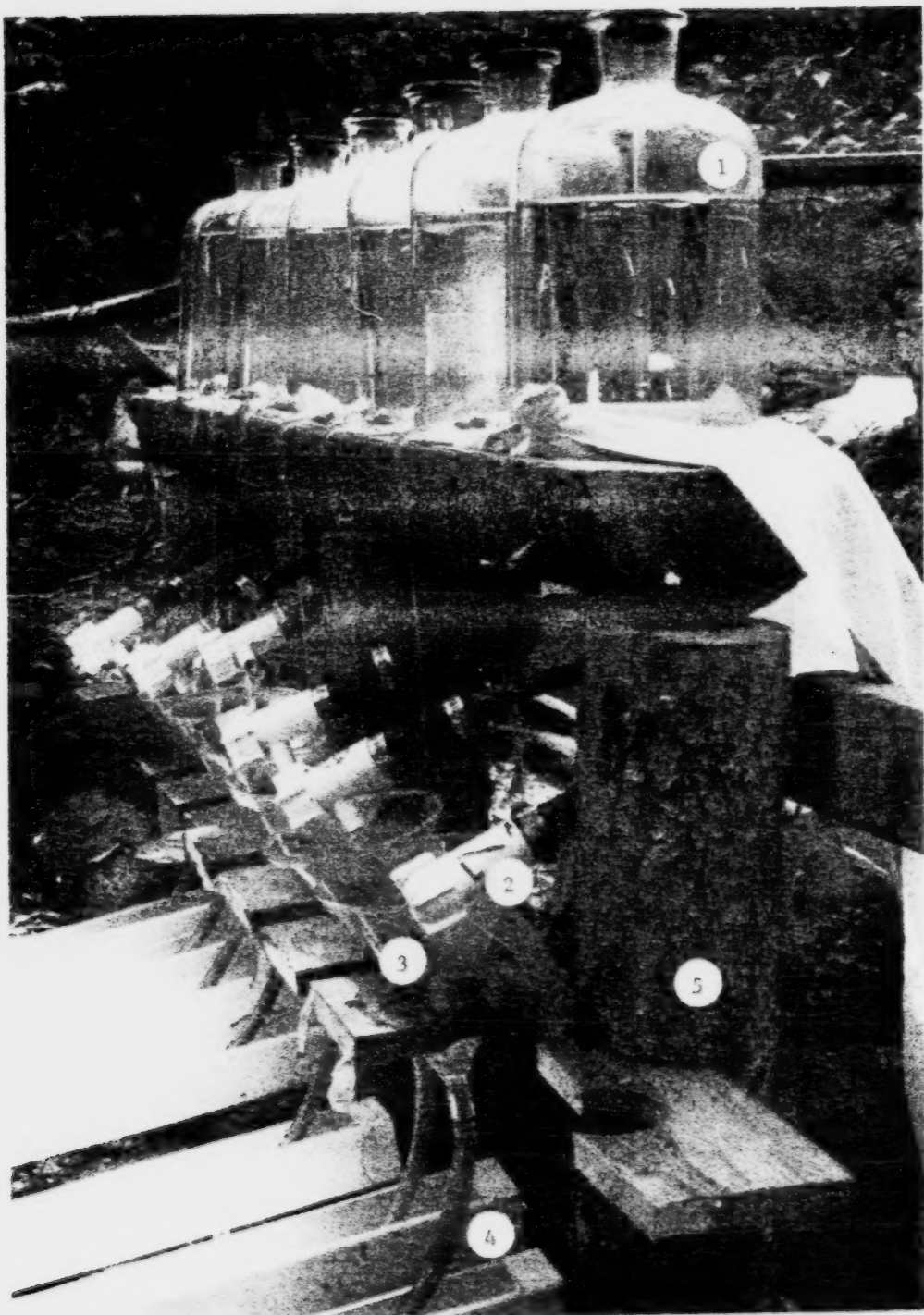
- A) blackflies (Simulium venustum), B) caddisflies (Pycnopsyche sp.), C) dragonflies (Ophiogomphus sp.), D) mayflies (Isonychia sp.), E) stoneflies (Lab: Phasganophora sp., Field: Acroneuria abnormis), F) crayfish (Orconectes propinquus).

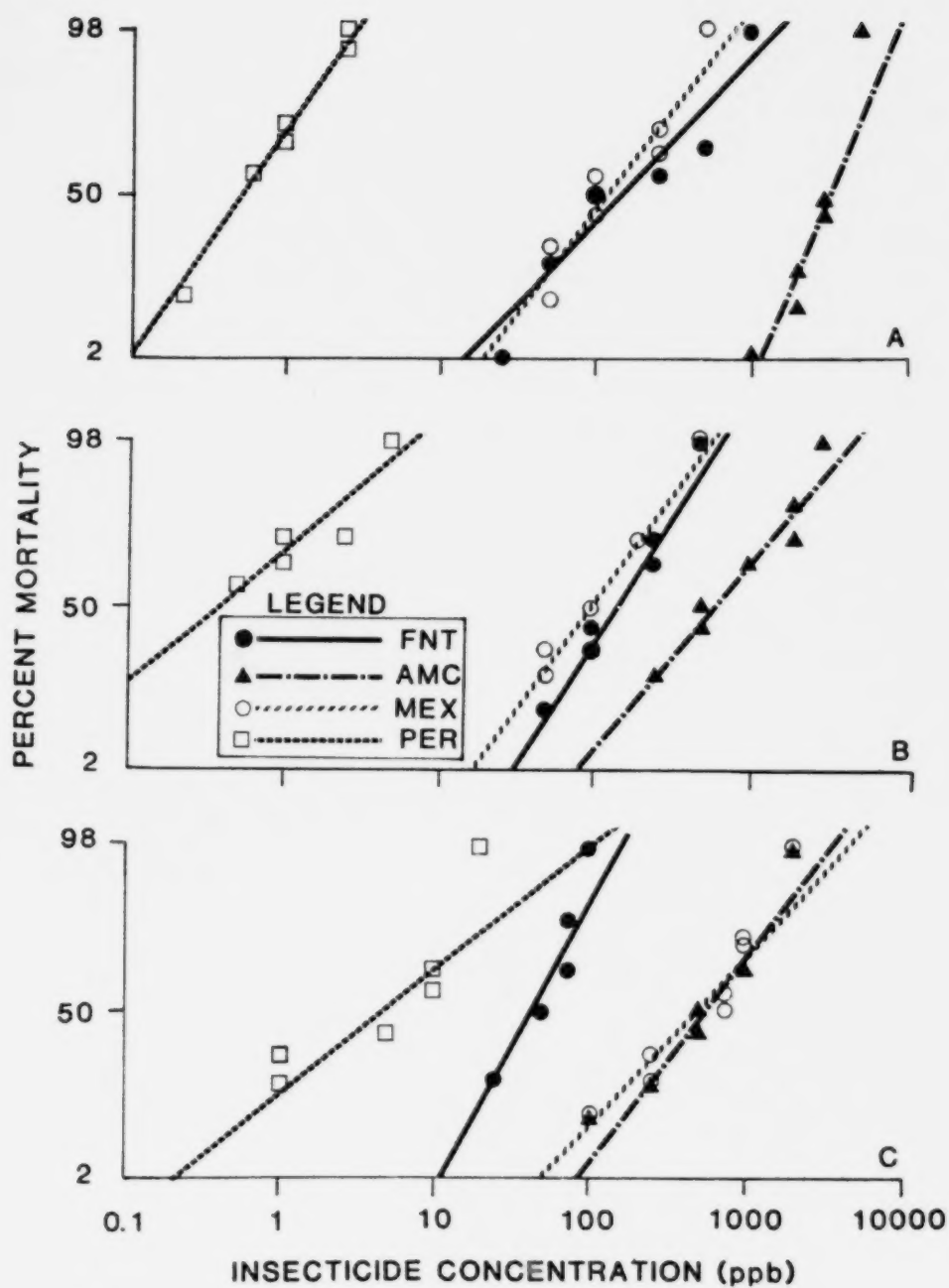
Figure 3: Fenitrothion concentration and aquatic invertebrate drift measured in Icewater Creek after an aerial application of fenitrothion at 280g A.I./Ha.

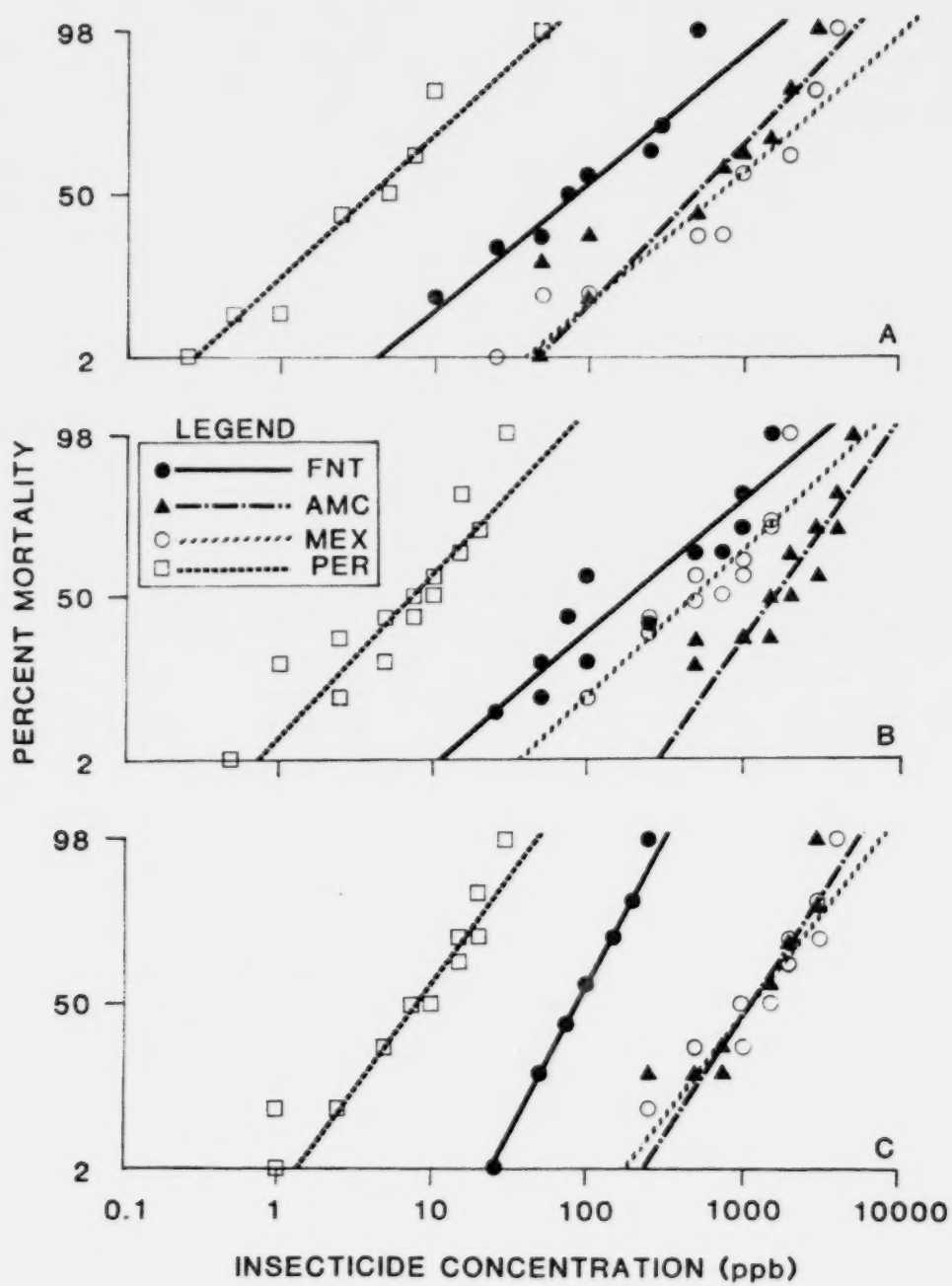
Figure 4: Comparisons between the toxicities of fenitrothion to aquatic invertebrates in laboratory, field and caged insect studies.

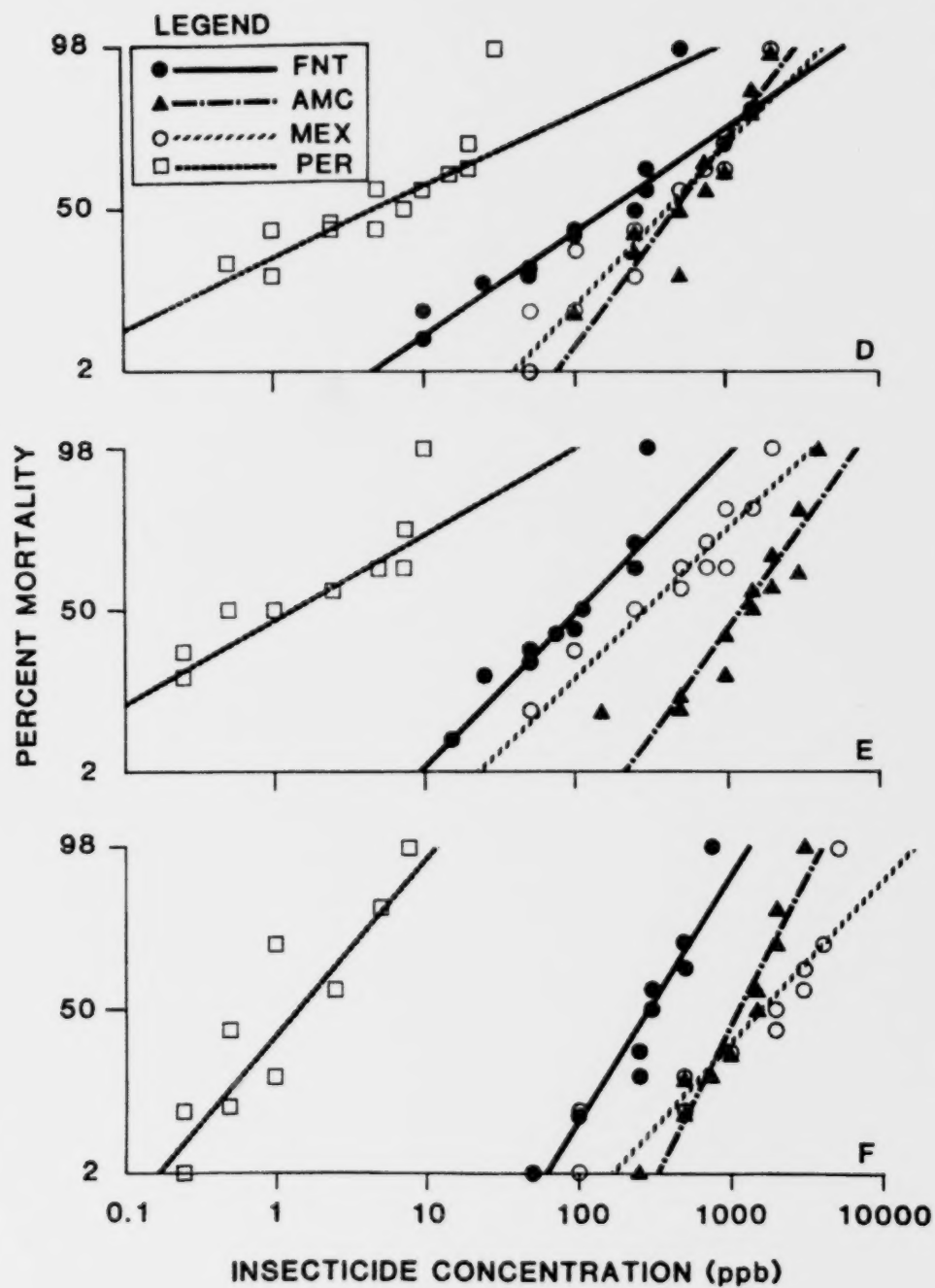
- A) blackflies (Simulium venustum), B) caddisflies (Pycnopsyche sp.), C) dragonflies (Ophiogomphus sp.), D) mayflies (Isonychia sp.), E) stoneflies (Lab: Phasganophora sp., Field: Acroneuria abnormis), F) crayfish (Orconectes propinquus).

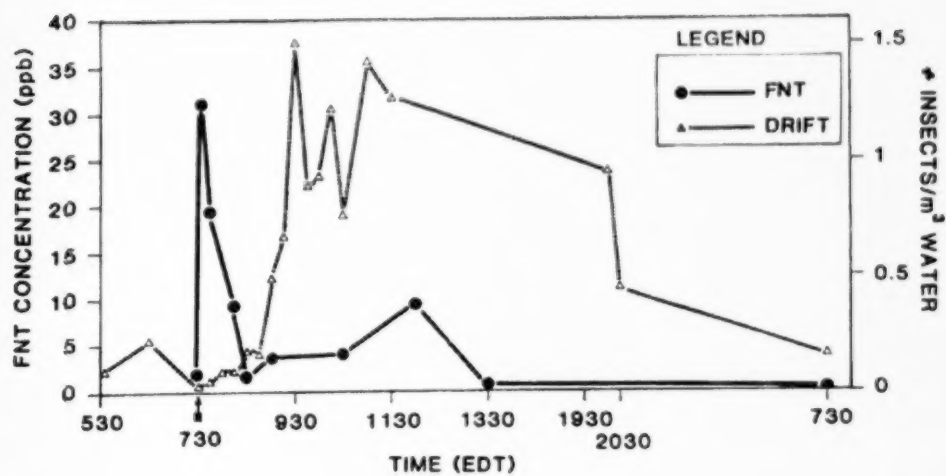












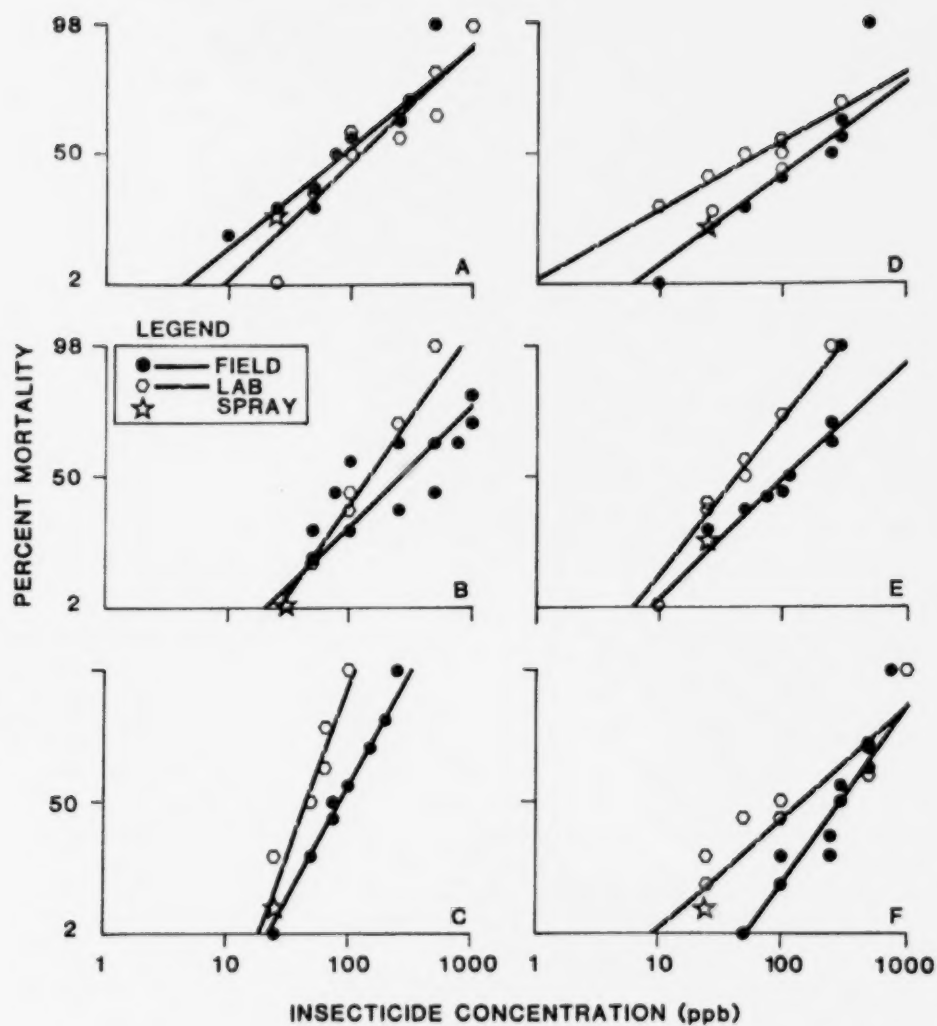


Table 1. Insecticides and formulations used in laboratory and field bioassays

Insecticide	Formulation	Class	A.I.	Emulsifier	Supplier
Permethrin	Ambush EC	Synthetic Pyrethroid	500 g/l	Incorporated	Chipman
Permethrin	Ambush ULV	Synthetic Pyrethroid	500 g/l	2% Acetone	Chipman
Permethrin	Micro- encapsulated	Synthetic Pyrethroid	200 g/l	N/A	Chipman
Fenitrothion	Sumithion Technical	Organo- Phosphate	94.6% w/w	1.1% Atlox 3409 F 1.1% dowanol	Sumitomo
Aminocarb	Matacil 180 D	Carbamate	19.6% w/w	2% Acetone	Chemagro
Aminocarb	Matacil 180 F	Carbamate	19.6% w/w	1.1% Atlox 3409 F	Chemagro
Mexacarbate	Zectran UCZF 19 Modified Batch 4 PLP-12-1	Carbamate	21.7% w/w	1.1% Triton X114	Union Carbide

Table 2. Habitats and trophic relationships⁽¹⁾ of aquatic invertebrates used in this study.

Taxa	Habitat	Habits	Trophic Relationships
Crustacea			
Astacidae			
<u>Orconectes propinquus</u>	Lotic-Erosional & Depositional	Crawlers	Herbivores Scavengers
Insecta			
Diptera			
Simuliidae			
<u>Simulium venustum</u>	Lotic-Erosional	Clingers	Filterers
Trichoptera			
Limnephilidae			
<u>Pycnopsyche</u> spp.	Lotic-Erosional & Depositional	Sprawlers Climbers	Scrappers Shredders, Chewers
Plecoptera			
Perlidae			
<u>Acroneuria abnormis</u>	Lotic, Lentic-Erosional	Clingers	Predators
<u>Phasganophora</u> spp.	Lotic-Erosional	Clingers	Predators
Ephemeroptera			
Oligoneuridae			
<u>Isonychia</u> spp.	Lotic-Erosional	Swimmers	Filterers, Predators
Odonata			
Gomphidae			
<u>Ophiogomphus</u> spp.	Lotic-Erosional & Depositional	Burrowers	Predators

1 - from Merritt and Cummins 1984.

Table 3. The toxicities of field formulations of four forest insecticides to selected aquatic invertebrates using a continuous flow laboratory bioassay apparatus.

	Ambush			Sumithion			Matacil 180F			Zectran UCZ19		
	Permethrin			Fenitrothion			Aminocarb			Mexacarbate		
	LC50	CL	Slope	LC50	CL	Slope	LC50	CL	Slope	LC50	CL	Slope
Blackflies (<i>Simulium</i>)	4.48 ^a	3.12 6.44	2.29 ^x	148.35 ^c	105.31 208.98	1.63 ^{wx}	2863.02 ^e	2582.48 3174.08	8.18 ^z	124.74 ^c	93.13 167.09	2.65 ^x
Caddisflies (<i>Pycnopsyche</i>)	3.17 ^a	1.56 6.45	1.19 ^w	136.52 ^c	104.49 178.36	3.01 ^{xy}	590.48 ^d	435.51 800.60	2.22 ^x	98.80 ^c	71.89 135.79	2.39 ^x
Dragonflies (<i>Ophiogomphus</i>)	4.37 ^a	2.54 7.52	1.32 ^{wx}	45.27 ^b	36.27 56.51	3.63 ^y	478.31 ^d	353.18 647.77	2.17 ^x	491.57 ^d	338.75 713.31	2.06 ^x

LC₅₀ values followed by the same letter are not significantly different at $p \leq 0.05$.

Slope values followed by the same letter are not significantly different at $p \leq 0.05$.

Table 4. The toxicities of field formulations of four forest insecticides to selected aquatic invertebrates using a continuous flow field bioassay apparatus.

	Ambush EC			Sumithion EC			Matacil 180F			Zectran UCZF19		
	Permethrin			Fenitrothion			Aminocarb			Mexacarbate		
	LC50	CL	Slope	LC50	CL	Slope	LC50	CL	Slope	LC50	CL	Slope
Blackflies (<u>Simulium</u>)	3.75 ^b	2.62 5.36	2.01 ^y	82.46 ^{cd} 131.42	51.74 131.42	1.55 ^y	344.43 ^{ef} 622.94	190.44 622.94	1.19 ^{xy}	718.67 ^f 1175.11	439.53 1175.11	1.32 ^y
Mayflies (<u>Isonychia</u>)	4.38 ^b	2.59 7.40	0.91 ^x	163.86 ^{cde} 232.57	115.46 232.57	1.38 ^y	478.93 ^f 618.11	371.10 618.11	2.24 ^y	356.39 ^f 492.57	257.86 492.57	1.75 ^y
Caddisflies (<u>Pycnopsycha</u>)	7.02 ^b	5.02 9.81	1.60 ^y	230.01 ^e 356.62	148.36 356.62	1.25 ^y	1275.60 ^g 1957.72	831.15 1957.72	1.17 ^{xy}	429.91 ^f 611.06	302.46 611.06	1.46 ^y
Stoneflies (<u>Acronuria</u>)	0.99 ^a	0.52 1.	0.85 ^x	96.51 ^{cd} 134.71	69.14 134.71	1.95 ^y	1062.12 ^g 1656.78	680.89 1656.78	1.24 ^{xy}	250.51 ^{ef} 366.07	171.44 366.07	1.51 ^y
Dragonflies (<u>Ophiogomphus</u>)	7.14 ^b	5.43 9.38	2.01 ^y	84.53 ^c 100.40	71.17 100.40	3.87 ^z	1016.64 ^g 1278.90	808.16 1278.90	2.74 ^{yz}	1049.55 ^g 1434.92	767.67 1434.92	1.88 ^y
Crayfish (<u>Orconectes</u>)	1.04 ^a	0.72 1.50	1.97 ^y	283.70 ^e 357.22	225.31 357.22	2.84 ^{yz}	1183.5 ^g 1433.23	977.28 1433.23	3.34 ^z	1503.65 ^g 2218.90	1018.96 2218.90	1.41 ^y

LC₅₀ values followed by the same letter are not significantly different at $p \leq 0.05$.

Slope values followed by the same letter are not significantly different at $p \leq 0.05$.

Table 5. Mortality of caged aquatic invertebrates after 24 hours exposure to fenitrothion; a) in Icewater Creek, and b) in a recirculating flow through laboratory bioassay apparatus after an experimental aerial spray program.

Insect	(a) % Mortality in Stream(1)	(b) % Mortality in Laboratory Bioassay Apparatus(2)
<u>Acroneuria</u>	13	40
<u>Isonychia</u>	11	20
<u>Simulium</u>	16	20
<u>Ophiogomphus</u>	5	10
<u>Pycnopsyche</u>	0	14
<u>Orconectes</u>	5	10

(1) For actual concentration of fenitrothion, see Figure 7.

(2) Fenitrothion concentration was 21.0 ppb and represents the average concentration over the 30 minute period when stream concentrations were at their peak.

SUMMARY - WORKSHOP ON NEEDS FOR CANADIAN ESTUARINE AND COASTAL WATER QUALITY GUIDELINES
P.G. Wells,² M.C. Taylor,¹ and A.R. Davis¹

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Panel Members: A.R. Davis,¹ A.R. Carlson,² L. Harding,³ R.C. Pierce,¹ M.C. Taylor,¹ and P.G. Wells⁴

The application of knowledge in marine ecotoxicology is essential for the conservation and protection of Canada's estuarine and coastal waters. One essential application is in the production of comprehensive water quality guidelines for these areas. In Canada, to date, such work has fallen between jurisdictions and, with the exception of the Shellfish Surveillance Area Guidelines, has been rare and fragmented.

The objective of this workshop was to review national and international work on development of guidelines and objectives, and discuss the need for generic guidelines for Canada. Water quality guidelines for freshwater which have been produced by a federal-provincial task force, are nearing completion and the first edition should be published in early 1987. This process of close cooperation among provincial and federal agencies may open the way to considering estuarine and coastal waters in the near future.

Main points of discussion:

- (1) The proposal for the development of guidelines was described by A.R. Davis. He requested the input of workshop participants and other aquatic toxicologists.
- (2) Margaret Taylor described the way in which guidelines are developed, and gave definitions of "criteria", "guidelines", "objectives", and "standards". Guidelines were defined as "numerical concentration or narrative statements recommended to support and maintain a designated water use" (COREM, in preparation). Important uses of estuarine and coastal waters include aquatic life, aquaculture, fisheries and recreation.
- (3) Wells, Carlson and Harding reviewed international and national activity, covering the U.K. (upgrading of estuaries, objectives for oxygen and specific chemicals), the EEC (objectives under Directive for Dangerous Substances), Australia, South Africa (water quality criteria for the South African coastal zone (1984)), the U.S.A. (Guidelines for Deriving Numerical National Water Quality Criteria for the Protection of Aquatic Organisms and Their Uses (1985); the 1980 and 1985 EPA series of Ambient Water Quality Criteria, for protection of aquatic life and its uses) and Canada (site- and problem-specific objectives on the east and west coasts, when problems or developments emerge). Other international activity is through UNEP where the "Montreal Guidelines" (1985) present two control strategies requiring marine water quality "standards".
- (4) An open discussion led to the following list of advantages and disadvantages of guidelines:

Estuarine and Marine Guidelines/Objectives - Canada

Advantages

1. Modify freshwater data
2. Clear definition of requirements
3. Leads to monitoring & assessment.
4. Good starting point.

Disadvantages

1. Need workable lists of parameters
2. Could be viewed as license to pollute.
3. Monitoring requirements - resources required.
4. Need to develop modelling expertise.
5. Need more exposure - recovery toxicological data to derive realistic objectives.
6. Need tests and protocol for specific marine organisms.

- (5) There was strong support for the estuarine and coastal guidelines approach at the Workshop, and a recognition that the recent UNEP "Montreal Guidelines" environmental protection framework was an ideal mechanism for responsible parties to discuss control strategies and instruments based on both use and environmental objectives. Setting objectives for estuarine and coastal water quality first requires having generic guidelines for the process and parameters involved, hence the need for federal and provincial departments to assist in developing the Canadian estuarine and coastal water quality guidelines.
- (6) The workshop ended on a note of optimism for this very clear application of the principles and data from aquatic toxicology.

Acknowledgements

A special thanks to the panel members and the participants who made the workshop a success. M. Gilbertson, J. Karau, and A. Chevrier participated in the continued workshop discussions. K. Thibault skillfully kept records of discussions. This summary was prepared with the support of Conservation and Protection, Environment Canada.

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RESUME - ATELIER SUR LE BESOIN DE RECOMMANDATIONS POUR LA QUALITE DES EAUX ESTUARINIENNES ET LITTORALES AU CANADA

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Membres du comité: A.R. Davis,¹ A.R. Carlson,² L. Harding,³ R.C. Pierce,¹ M.C. Taylor,¹ et P.G. Wells⁴

L'application des connaissances en écotoxicologie marine est essentielle afin d'assurer la conservation et la protection des eaux estuariennes et littorales du Canada. L'élaboration de recommandations détaillées pour la qualité de l'eau pour ces lieux sert d'application essentielle. Jusqu'à présent, au Canada, la responsabilité pour cet ouvrage a tombé entre diverses juridictions; de plus, les ouvrages disponibles sont rares et fragmentaires.

Le but de cet atelier était de passer en revue les ouvrages internationaux et nationaux sur les recommandations et les objectifs pour la qualité des eaux ainsi que de discuter des besoins pour des recommandations génériques pour la qualité de l'eau au Canada. Les recommandations pour la qualité de l'eau douce, élaborées par un groupe de travail fédéral-provincial, sont près d'être achevées et paraîtront au début de l'année 1987. Ce processus de coopération étroite entre les organismes provinciaux et fédéraux amorcés peut-être des réflexions en matière des eaux estuariennes et littorales dans un avenir rapproché.

Principaux points de la discussion:

- (1) La proposition pour l'élaboration des recommandations fut décrite par A.R. Davis. Il invita les commentaires des participants à l'atelier ainsi que de d'autres toxicologues aquatiques.
- (2) Margaret Taylor a décrit les démarches suivies lors de l'élaboration des recommandations et elle a donné la définition de "critères", "recommandations", "objectifs" et "normes". On entend par ces recommandations: des "énoncés ou valeurs numériques limites de la concentration recommandés pour subvenir à une utilisation donnée de l'eau et la maintenir" (COMÉ, en préparation). Les usages importants des eaux estuariennes et littorales comprennent la vie aquatique, l'aquaculture, les pêches et les fins récréatives.
- (3) Wells, Carlson et Harding ont revu les activités internationales et nationales, y compris celles de la Grande Bretagne (amélioration des estuaires, objectifs pour l'oxygène et pour des produits chimiques spécifiques), de la CEE (objectifs d'après la "Directive for Dangerous Substances"), de l'Australie, de l'Afrique du Sud (Water quality criteria for the South African coastal zone (1984)), des E.-U. ("Guidelines for Deriving Numerical National Water Quality Criteria for the Protection of Aquatic Organisms and Their Uses (1985)"; la série 1980 et 1985 de "Ambient Water Quality Criteria" de l'EPA visant la protection de la vie aquatique et ces usages) et du Canada (des objectifs pour des lieux et des problèmes spécifiques pour les côtes de l'est et de l'ouest, lorsque des problèmes ou des développements surgissent). D'autres activités internationales découlent de l'UNEP dont les "Montreal Guidelines" (1985) développent deux stratégies de contrôle exigeant des "normes" de qualité des eaux marines.
- (4) Un échange ouvert a mené à l'énumération d'avantages et de désavantages des recommandations ci-dessous:

Recommandations pour la qualité des eaux estuariennes et littorales - Canada

Avantages

1. Modifier les données d'eau douce
2. Définition claire des exigences
3. Mène au monitoring et à l'évaluation
4. Bon point de départ

Désavantages

1. Besoin d'une liste réalisable de paramètres
2. Susceptibles d'être perçus comme accordant la liberté de polluer
3. Exigences du monitoring - ressources exigées
4. Besoin de développer des compétences dans l'élaboration de modèles
5. Besoin davantage de données toxicologiques sur l'exposition - effect afin d'en extraire des objectifs réalisables
6. Besoin d'essais et de protocoles pour des organismes marins

- (5) A l'atelier, l'approche des recommandations pour les eaux estuariennes et littorales a reçu un appui vivement favorable. On reconnaît également que dans le cadre de la protection environnementale, tel que décrit dans les récents "Montreal Guidelines" de l'UNEP, un processus idéal se présente afin d'entamer, après des intervenants responsables, des discussions en matière de stratégies de contrôle basées sur l'utilisation de même que sur les objectifs environnementaux. L'élaboration d'objectifs pour la qualité des eaux estuariennes et littorales exige d'abord la disponibilité de recommandations génériques pour le processus et pour les paramètres en question, d'où le besoin d'une collaboration parmi les ministères fédéraux et provinciaux lors de l'élaboration de recommandations pour la qualité des eaux estuariennes et littorales au Canada.
- (6) L'atelier termine sur une note optimiste en matière de cette application très claire des principes et des données de la toxicologie aquatique.

Remerciements

On remercie particulièrement les membres du comité ainsi que les participants qui ont fait de l'atelier un succès. M. Gilbertson, J. Karau et A. Chevrier ont participé, le lendemain, à la reprise des discussions de l'atelier. On apprécie les renseignements et les réflexions contribués par J. Alabaster (C.B.), D. Valiela (DQI, Vanc.) et P. Eaton (SPE, Dart.). K. Thibault a tenu avec habileté le procès-verbal des discussions. Ce résumé a été rédigé avec le soutien de Conservation et protection, Environnement Canada.

Sea Urchin Sperm Bioassay For Measuring Sewage Treatment
Efficiency and Potential Toxicity in Marine Waters

by

Paul A. Dinnel and Quentin J. Stober

Abstract

A newly standardized sea urchin sperm bioassay protocol was used to measure the toxicity of sewage at each stage of the treatment process to assess removal of sewage components causing acute toxicity to sensitive marine animals. The sea urchin sperm bioassay is a rapid test procedure that exposes seawater-activated sperm cells in vitro to test or control solutions for 60 minutes. Eggs are then added and time allowed for fertilization to take place followed by fixation with formalin. The degree of fertilization success is determined by noting the presence or absence of the vitelline (fertilization) membrane in subsamples of eggs. Ten grab samples of each stage of sewage (with appropriate controls) were tested during both summer and winter for two successive years using eleven sewage dilutions (0.27 to 20.0% vol/vol in seawater). Chlorinated secondary sewage was most toxic followed in descending order to toxicity by influent, primary, dechlorinated secondary and unchlorinated secondary sewage. Primary treatment afforded only a slight reduction in acute toxicity over that observed for influent sewage while secondary treatment markedly reduced toxicity. Unchlorinated and chlorinated-dechlorinated secondary sewage were essentially equal in toxicity. A high degree of variability in toxicity was found between sampling days and between

years. Additionally, three other sensitive marine animal life stages were bioassayed side-by-side with the sperm assays. The results showed that the 60-min sperm assay was as sensitive as a 96-hr sea urchin embryo development assay, slightly more sensitive than a 48-hr Pacific oyster embryo development assay and substantially more sensitive than a 48-hr Dungeness crab zoea assay. The results of this study show that (1) a simple 60-min sperm bioassay procedure is quicker and at least as sensitive as 48 to 96-hr embryo or larval bioassays for measuring toxicities of complex sewage effluents, (2) a sperm assay can be used to trace the efficiency of the sewage treatment process relative to acute toxicity in marine waters, (3) that secondary treatment is an important step in the treatment process for removing acute toxicity prior to discharge and (4) that data of this type can be used to determine effluent dilution rates necessary to avoid acute toxicity to sensitive marine animal life stages.

Paul A. Dinnel and Quentin J. Stober

RESUME

Un nouveau protocole standardisé du test biologique du sperme d'oursin a été utilisé pour mesurer la toxicité des eaux résiduaires à chaque étape du processus de traitement servant à évaluer l'enlèvement des composants résiduaires causant une toxicité aiguë pour les animaux marins qui y sont sensibles. Les procédures du test biologique du sperme d'oursin sont rapides et exposent l'eau de mer avec les cellules spermatiques activées in-vitro pour tester ou contrôler les solutions durant une période de 60 minutes. Les oeufs sont alors ajoutés et le temps est alloué pour que s'effectue la fertilisation vivie par la fixation avec du "formalin". Le degré de fertilisation est déterminé par l'observation de la présence ou absence de la membrane vitelline parmi des échantillons d'oeufs. Deux prélèvements d'échantillons de chaque étape résiduaires (avec contrôles appropriés) ont été testés l'été et l'hiver durant deux années successives utilisant 11 dilutions d'eaux résiduaires (0.27 à 20.0% vol/vol. d'eau de mer.). Les eaux résiduaires secondaires chlorées étaient les plus toxiques suivies en ordre décroissant de toxicité par, premièrement, l'affluence, deuxièmement, par la déchloration et par les eaux résiduaires secondaires non-chlorées. Le premier traitement procurait seulement une mince réduction de la toxicité critique d'après ce qui a été observé lors de l'affluence des eaux résiduaires alors que le second traitement réduisait, d'une façon prononcée, la toxicité. Les eaux résiduaires secondaires non-chlorées et chlorées/dechlorées étaient fondamentalement égales en toxicité. Un haut niveau de variation dans la toxicité a été détecté entre les journées d'échantillonnage et les années. En plus, 3 autres stades sensibles dans la vie d'animaux marins ont été testés biologiquement côte-à-côte avec les tests spermatiques. Les résultats démontraient que le test étalé sur 60 min. était, aussi sensible que le test sur le développement embryonique de l'oursin étalé sur 96 heures, légèrement plus sensible que le test de 48 hr. effectué sur le développement embryonique des huitres du Pacifique, et substantiellement plus sensible qu'un test de 48 hr. sur les zoes du crabe "Dungeness". Les résultats de cette étude montrent que (1) la procédure d'un simple test biologique du sperme d'une durée de 60 min. est plus rapide et au-moins aussi sensible que les tests embryoniques et larvaires d'une période de 48 et 96 hr pour le mesurage des toxicités des effluents d'eaux résiduaires ayant une certaine complexité, (2) un test spermatique peut être utilisé pour déterminer l'efficacité des processus de traitement des eaux résiduaires relatifs à la toxicité critique dans les eaux marines, (3) le deuxième traitement est une étape importante dans le processus de traitement pour l'enlèvement de la toxicité critique précédant le déversement et (4) les données de ce type peuvent être utilisées pour déterminer le taux de dilution des effluents, nécessaire pour éviter une toxicité critique aux différents stades sensibles du développement des animaux marins.

DEMARCHE ECOTOXICOLOGIQUE POUR EVALUER
LA TOXICITE DE SEDIMENTS MARINS A DRAGUER

ECOTOXICOLOGICAL APPROACH TO EVALUATE
THE HAZARD OF MARINE DREDGED SEDIMENTS

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RÉSUMÉ

Les sédiments marins dragués à proximité des ports et des agglomérations contiennent souvent des contaminants en concentrations excessives; en conséquence, leur immersion peut constituer un risque pour la faune et la flore. L'évaluation de ce risque exige non seulement l'analyse chimique des sédiments à draguer, mais aussi la mesure de leur écotoxicité. Or, les essais biologiques permettant de mesurer cette écotoxicité exigent des longues durées (un à six mois), des installations complexes et/ou des sédiments extrêmement toxiques.

Afin de pouvoir effectuer des essais biologiques plus simples et plus rapides, comme le souhaitent les gestionnaires de l'environnement, nous avons envisagé les procédés d'élutriation, de lixiviation et de diffusion.

Seul le procédé de lixiviation a permis, en reproduisant des conditions représentatives du milieu, de mesurer l'écotoxicité de sédiments provenant surtout du port d'Halifax. Ces sédiments contenaient des métaux lourds, des cyanures, des huiles et des BPC. Au cours de ce procédé, qui a duré 24 heures, nous avons utilisé un mélange sédiments-eau dans une proportion de 1:10 et obtenu un lixiviat ayant des teneurs en métaux lourds et en huiles plus élevées que les normes de qualité du milieu marin.

Nous avons mesuré l'écotoxicité du lixiviat à trois niveaux, soit l'écotoxicité létale, sublétale et chronique, lesquels correspondent à trois niveaux de protection environnementale dans une "spirale d'écotoxicité".

Aucun effet létal ne fut observé chez les moules Mytilus edulis après quinze (15) jours d'exposition au lixiviat. Le lixiviat avait toutefois un effet sublétal sur les diatomées Thalassiosira pseudonana, inhibant faiblement leur croissance pendant huit (8) jours; cet effet ne pouvait être discerné qu'après avoir fait le décompte de la stimulation de croissance imputable aux NO_3^- et NH_4^+ contenus dans les sédiments du lixiviat. Par ailleurs, on n'obtint aucune indication avec deux autres essais biologiques plus rapides en vue d'établir la sublétalité, à savoir celui de la bioluminescence (5 minutes) des bactéries Photobacterium phosphoreum et celui de la mobilité (24 heures) des microcrustacés Artemia salina. Par contre, le potentiel de toxicité chronique du lixiviat a été mis en évidence au moyen de deux essais biologiques relativement simples et rapides; en effet, on a observé une mutagénicité chez les bactéries Salmonella typhimurium en trois jours et un début de bioaccumulation des métaux lourds après sept jours chez les moules Mytilus edulis.

RÉSUMÉ
(suite)

Bref, sauf dans le cas de sédiments extrêmement contaminés, la biodisponibilité des contaminants dans les sédiments marins à draguer est faible et leur effet écotoxique est lent. Malgré ceci, leur potentiel d'écotoxicité chronique et le début de leur effet subléthal se sont révélés décelables au moyen d'essais biologiques simples et rapides (8 jours) après lixiviation.

SUMMARY

Marine sediments dredged in the vicinity of harbours and urban areas often contain contaminants in excessive concentrations. Dumping these sediments in the ocean can therefore represent a threat to aquatic life. The evaluation of this threat requires a chemical analysis of sediments to be dredged, as well as a measure of their ecotoxicity. The bioassays available for this evaluation require time (from one to six months) and complex experiences, or can only detect toxicity at very high levels.

In an effort to formulate simpler and faster bioassays, as required by environmental managers, three procedures have been evaluated in this report : elutriation, lixiviation and diffusion.

Only the lixiviation bioassay permitted a measurement of the toxicity of sediments dredged mostly from the Halifax harbour, while recreating representative conditions. These sediments contained heavy metals, cyanides, oils and PCBs. In the course of the bioassay, which lasted 24 hours, a mixture of sediments and water in a ratio of 1:10 was used and the lixivate obtained had a higher dissolved content of heavy metals and oils than allowed by sea water quality norms.

The ecotoxicity of the lixivate was measured at three levels, lethal, sublethal and chronic, corresponding to three levels of environmental protection in an "ecotoxicity spiral".

No lethal effect was observed on Mytilus edulis mussels after 15 days of exposure to the lixivate. The lixivate had a sublethal effect on Thalassiosira pseudonana diatoms, slightly inhibiting their growth during eight days; this latter effect could only be detected after having discounted growth stimulation owed to NO_3^- and NH_4^+ contained in the lixivate sediments. However, no indication was obtained with two other faster bioassays to determine sublethality, i.e. bioluminescence (5 minutes) of Photobacterium phosphoreum bacteria and mobility (24 hours) of Artemia salina microcrustaceans. Despite that, the chronic toxicity potential of the lixivate was evidenced through two relatively simple and quick bioassays. Mutagenicity was observed on Salmonella typhimurium bacteria after three days and an incipient bioaccumulation of heavy metals was observed after seven days on Mytilus edulis mussels.

In summary, except in the case of highly contaminated sediments, there is little bioavailability of contaminants in the sediments to be dredged and their ecotoxicity effect is slow. However, their chronic ecotoxicity potential and their incipient sublethal effect were detectable through simple and quick bioassays (8 days) after lixiviation.

1. INTRODUCTION

Les activités de dragage essentielles à la circulation navale déplacent près de 20 millions de m³ de sédiments par année le long des côtes canadiennes (Levings, 1983). Dans plusieurs cas, les sédiments dragués se trouvent à proximité de ports et d'agglomérations industrielles et contiennent alors plusieurs contaminants résultant de l'activité humaine. Par exemple, au port d'Halifax, on drague annuellement de 15 000 à 30 000 m³ de sédiments qui contiennent notamment en moyenne 425 mg/kg de Pb et 1320 mg/kg de BPC (Levings, 1983).

Lorsque ces sédiments, une fois dragués, sont rejetés à la mer, les contaminants qu'ils renferment risquent de nuire à la faune et la flore dans le voisinage du lieu de rejet.

Ce risque préoccupe les gestionnaires de l'environnement qui ont besoin, pour l'évaluer, de données déterminantes acquises au moyen de méthodes relativement rapides et simples.

2. PROBLÉMATIQUE

Pour répondre à ce besoin, il existe deux solutions complémentaires : l'analyse des principaux contaminants présents dans les sédiments à draguer et des essais biologiques mesurant la toxicité de ces sédiments.

La première est couramment appliquée et des normes de qualité sont en vigueur depuis 1975 au Canada (voir tableau 1).

Par contre, la seconde s'avère plus complexe et a fait l'objet de divers types d'expérimentation (voir tableau 2).

TABLEAU 1

NORMES CANADIENNES RELATIVES AUX CONTAMINANTS
DANS LES SÉDIMENTS MARINS DE DRAGAGE*

<u>CONTAMINANTS</u>		<u>NORMES*</u>
		(en mg/kg en poids sec)
a)	<u>Substances interdites</u>	
	. Organochlorés	
	BPC	0,05
	Chlordane	"
	DDT et dérivés	"
	Dieldrine et analogues	"
	HCB	"
	Autres	"
	. Métaux lourds	
	Cadmium	0,6
	Mercure	0,75
	. Huiles et graisses	10,0
b)	<u>Substances réglementées</u>	
	. Métaux lourds	
	Chrome	100
	Cuivre	48
	Nickel	55
	Plomb	20
	Zinc	80
	. Autres inorganiques	
	Arsenic	3
	Cyanures	0,1

* Source : Environnement Canada, 1975

TABLEAU 2

REVUE DES PRINCIPAUX TYPES D'ESSAIS BIOLOGIQUES

FAITS AVEC DES SÉDIMENTS MARINS

<u>PARAMÈTRES ÉTUDIÉS</u>	<u>ORGANISMES TESTÉS</u>	<u>DURÉES DES TESTS</u>	<u>RÉFÉRENCES</u>
a) <u>Bioaccumulation</u>			
Bioaccumulation de BPC ajoutées à des sédiments	Ver : <u>Nereis deversicolor</u>	60 jours	Elder et coll., 1979
Bioaccumulation de BPC combinés à des sédiments	Ver : <u>Nereis virens</u>	32 jours	McLeese et coll., 1980
Bioaccumulation de BPC des sédiments du port d'Halifax (N.B. : leurs métaux lourds n'étaient guère accumulés)	Bivalve : <u>Macoma balthica</u> (tissus mous)	32 jours	EPS, 1980
Bioaccumulation de Cd des sédiments proches de Dalhousie, Nouveau-Brunswick	Bivalve : <u>Macoma balthica</u> (tissus durs)	20 jours	MacLaren Marex, 1980
Bioaccumulation de BPC, Hg, Pb, des sédiments du port de Vancouver	Bivalves : <u>Macoma balthica</u> et	30 jours	McGreer et coll., 1981
	<u>Mytilus edulis</u> (tissus mous)	60 jours	McGreer et Reid, 1983
Bioaccumulation de BPC ajoutées à des sédiments	Crevette : <u>Crangon septemspinosa</u>	32 jours	McLeese et coll., 1980
b) <u>Létalité</u>			
Létalité faible des sédiments du port d'Halifax	Bivalve : <u>Macoma balthica</u>	32 jours	EPS, 1980
Létalité de sédiments auxquels 8 organo-chlorés étaient ajoutés	Crevette : <u>Crangon septemspinosa</u>	4 jours	McLeese et Metcalfe, 1980

TABLEAU 2

REVUE DES PRINCIPAUX TYPES D'ESSAIS BIOLOGIQUES

FAITS AVEC DES SÉDIMENTS MARINS

(suite)

<u>PARAMÈTRES ÉTUDIÉS</u>	<u>ORGANISMES TESTÉS</u>	<u>DURÉES DES TESTS</u>	<u>RÉFÉRENCES</u>
Létalité de sédiments du port de Baltimore	Poisson : <u>Fundulus heteroclitus</u>	1 jour	Tsai et coll., 1979
Létalité des suspen- sions de sédiments provenant d'une drague à Puget Sound, Washington	Poisson : <u>Oncorhynchus keta</u>	4-7 jours	Smith, 1978
Létalité des suspen- sions de sédiments en concentrations élevées	Divers : bivalve, crevette, crabe, poissons	2-7 jours	Peddicord et McFarland, 1978
Létalité après lixi- viation des sédiments proches des rejets d'une usine de pâtes et papiers à Inlet Albertini, Colombie- Britannique	Crevette : <u>Crangon septemspinosa</u>	2-4 jours	EVS Consultants, 1977
c) <u>Sublétalité</u>			
Enfouissement dans des sédiments de l'estuaire du fleuve Fraser et du port de Vancouver	Bivalve : <u>Macoma balthica</u>	2 jours	McGreer, 1979 McGreer et coll., 1981
Évitement pour des sédiments contaminés en suspension	Poisson : <u>Clupea harengus</u>	1 jour	Wildish et coll., 1977
Consommation d'oxy- gène en présence de sédiments contaminés	Poisson : <u>Morone saxatilis</u>	1 jour	O'Connor et coll., 1977

Si l'on considère les divers essais biologiques qui ont été effectués avec des sédiments marins, on constate que, de façon générale, ils exigent de longs délais et/ou des installations spéciales. Des essais biologiques plus rapides et/ou plus simples sont souhaités par les gestionnaires de l'environnement pour mesurer la toxicité des sédiments marins.

En vue de répondre à cette attente, on peut faire appel à des procédés de laboratoire qui activent à court terme des phénomènes survenant à plus long terme dans le milieu. Trois possibilités sont offertes.

- i) L'élutriation consiste à mélanger vigoureusement les sédiments avec de l'eau pendant 30 minutes (proportion 1:4; actuellement, on tend vers 1:20 selon Rochon, 1984), à laisser décanter durant 60 minutes et à recueillir l'élutriat (phase aqueuse à filtrer) pour des tests de toxicité (U.S. Army Corps of Engineers, 1976). Cette méthode vise à simuler un déversement de sédiments pris par une drague suceuse. En eau douce, elle favorise une libération et ainsi une biodisponibilité des métaux lourds et des organochlorés présents dans les sédiments. Par contre, en milieu marin, ceci ne semble pas survenir (EPS, 1976) sauf à pH très basiques, tels que le pH 10 pour le mercure (Murakami et Tokeishi, 1977) ou à des pH très acides, tels que les pH 2-3 pour les autres métaux lourds (Levings, 1983). Ces pH n'étant guère représentatifs pour des études environnementales, la méthode d'élutriation est peu recommandée pour les sédiments marins.
- ii) Pour la lixiviation, on mélange également les sédiments et l'eau, mais les conditions sont moins sévères que pour l'élutriation : proportion similaire pour les sédiments et

l'eau (1:4 à 1:20) mais agitation plus modérée (trois rotations par minute) et durée plus longue (quelques heures à quelques jours) (WTC, 1981; Côté et Constable, 1982). Après décantation, le lixiviat (phase aqueuse) contient généralement une partie appréciable des contaminants qui étaient associés aux sédiments et fait alors l'objet de tests de toxicité.

- iii) La diffusion des sédiments en suspension dans des bacs peut également être une façon d'étudier les effets de l'immersion de sédiments en considérant les conditions de courant. Pour obtenir cette diffusion, on utilise un courant continu (0,2 l/min) en circuit fermé avec des suspensions de sédiments en proportion 1:4 à 1:20 (Thellen et coll., 1984).

En optant pour les procédés par lixiviation et par diffusion, on peut escompter activer la libération des contaminants contenus dans les sédiments et, subséquemment, leur écotoxicité et ainsi simuler en peu de temps des libérations et des écotoxicités analogues à celles qui surviendraient lors des dragage, charriage, immersion et diffusion des sédiments marins.

3. PROVENANCE ET ANALYSE DES SÉDIMENTS

Pour expérimenter les deux solutions retenues dans la problématique, il fallait d'abord disposer de sédiments marins suffisamment contaminés. On a d'abord choisi ceux du port d'Halifax vu que des analyses faites en 1982 avaient montré que leurs teneurs en métaux lourds, en huiles et en BPC étaient très élevées (Levings, 1983). Ils ont été prélevés le 15 novembre 1983 à 10 m de la jetée n° 2 du port, à une profondeur inférieure à 20 m; 10 kg furent recueillis avec une benne Ponar.

Immédiatement après leur prélèvement, les sédiments furent placés et congelés dans des sacs en plastique pour envoi ultérieur au laboratoire.

Leur analyse physico-chimique a été faite selon des techniques courantes :

eau	séchage à 80°C pendant 24 heures
huiles et graisses	EPA, 1979
C organique	APHA et coll., 1980
métaux lourds	BEST, 1981 et contrôles - standards**
mercure	BEST, 1979
cyanure	APHA et coll., 1980
BPC* et autres	
organochlorés	BEST, 1980 et contrôles - standards**
granulométrie	Walton, 1980

Les résultats de cette analyse sont exposés au tableau 3. Ils indiquent que les sédiments recueillis avaient, par rapport aux normes précitées (voir tableau 1), des teneurs nettement excessives en métaux lourds (sauf Cr), en cyanures et en huiles et pouvaient de ce fait être considérés comme très contaminés bien que leurs concentrations en organochlorés soient inférieures à ces normes (sauf pour les BPC en teneurs quelque peu élevées).

Ces sédiments avaient une texture essentiellement limoneuse et un pourcentage appréciable de carbone organique; ces deux facteurs favorisent l'adsorption des contaminants (Levings, 1983).

* BPC analysés : Arochlore 1254 et 1260.

** Les contrôles avec des standards ont permis de vérifier la fiabilité des résultats : la variation moyenne obtenue ne dépassait pas 12%.

TABLEAU 3

CARACTÉRISTIQUES PHYSICO-CHIMIQUES
DES SÉDIMENTS TESTÉS

<u>Paramètres</u>	<u>Concentrations</u> (en mg/kg en poids sec)
	<u>Proportions</u> (en pourcentages)
Eau	55 %
<u>Métaux lourds :</u>	
Cd	4
Cr	44
Cu	150
Hg	2
Pb	560
Zn	610
Cyanures	4
C organique	4 %
Huiles et graisses	730
<u>Organochlorés :</u>	
BPC	0,16
Chlordane	$< 35,0 \times 10^{-3}$
DDT	$< 35,0$ "
HCB	$< 10,0$ "
<u>Granulométrie :</u>	
Sable grossier (0,25 μm)	0 %
Sable fin (0,06 - 0,25 μm)	7 %
Limon (0,002 - 0,06 μm)	70 %
Argile (0,002 μm)	23 %

N.B. : Analyses faites en triplicata

Leur cinétique de sédimentation a été examinée en vue d'évaluer à quelle vitesse ils se déposeraient et se disperseraient en fonction des courants après avoir été dragués et immergés. Pour cela, on mélangea sédiments et eau en proportion 1:25 dans un cylindre d'un litre et, après une agitation destinée à assurer l'homogénéité de la suspension, on préleva des échantillons de 25 ml aux 3/10^{èmes} du cylindre à différents intervalles de temps : la concentration de solides en suspension dans ces échantillons était ensuite déterminée après séchage de 24 h à 100° C. Ce test indiqua que les sédiments s'étaient vite déposés (96, 98 et 99,5% après respectivement 1, 4 et 24 heures).

4. LIXIVIATION DES SÉDIMENTS

Le procédé de lixiviation décrit plus haut (voir paragraphe 2) a été appliqué dans les conditions suivantes :

- . Proportion sédiments-eau : 1:10
- . Eau de mer synthétique : formulation commerciale "Forty Fathoms" dissoute dans de l'eau déminéralisée à raison de 33 g/l pour obtenir une salinité de $2,7 \pm 0,05\%$ avec pH $7,5 \pm 0,3$ et ensuite "maturée" pendant 24 heures avec une lente agitation; mentionnons ici que, dans le contexte de la recherche d'une méthode simple et standardisable, l'emploi d'une eau marine synthétique est plus commode et adéquat que le pompage, le transport et la préservation d'une eau marine prise à un site;
- . Mélange par agitation rotative : 3 révolutions par minute
- . Aucun ajustement de pH

- . Durée : 24 heures
- . Filtration (0,45 μ m) finale du lixiviat

Suite à la lixiviation, la composition chimique des sédiments du port d'Halifax avait changé. Leurs teneurs en cyanures, en C organique et en huiles avaient diminué respectivement 2, 4 et 7 fois. Par contre, leurs teneurs en métaux lourds et en organochlorés restaient relativement les mêmes (15% de variation).

L'eau du lixiviat, analysée au moyen de techniques courantes (les références à ce sujet correspondent à celles citées au paragraphe 3), contenait plusieurs des contaminants présents dans les sédiments. Sa qualité ne respectait pas les normes propices à la vie aquatique marine, notamment pour les métaux lourds globalement et les huiles, tel qu'indiqué au tableau 4.

On pouvait donc s'attendre à ce que le lixiviat des sédiments du port d'Halifax occasionne une écotoxicité.

5. ÉCOTOXICITÉ DU LIXIVIAT DES SÉDIMENTS

Pour tester l'écotoxicité d'un rejet en milieu aquatique, une multitude d'essais biologiques sont proposés. De façon générale, le choix des essais biologiques est fondé sur les exigences suivantes : représentativité des organismes, sensibilité, simplicité des tests, rapidité des réponses, "reproductibilité" des résultats et coût raisonnable.

Aucun essai biologique ne pouvant satisfaire à toutes ces exigences à la fois, on préconise de plus en plus la combinaison de quelques essais biologiques de type différent. En outre,

TABLEAU 4

CARACTÉRISTIQUES PHYSICO-CHIMIQUES
DES LIXIVIATS DES SÉDIMENTS TESTÉS

<u>Paramètres</u>	<u>Concentrations</u> (en mg/l)	<u>Normes de qualité</u> <u>pour la vie aquatique</u> <u>marine*</u> (en mg/l)
<u>Métaux lourds :</u>		
Cd	0,06	0,005
Cr	0,05	0,005
Cu	0,10	0,05
Hg	0,001	0,0001
Pb	0,35	0,05
Zn	0,28	0,10
Cyanures	0,003	0,005
Huiles et graisses	190	0,01 à 0,1
<u>Organochlorés :</u>		
BPC	< 0,09 X 10 ⁻³	0,001 X 10 ⁻³
Chlordane	< 0,02 X 10 ⁻³	0,004 X 10 ⁻³
DDT	< 0,005 X 10 ⁻³	0,001 X 10 ⁻³
HCB	< 0,005 X 10 ⁻³	0,004 X 10 ⁻³
Salinité	2,9 ‰	
pH	8,1	6,5 à 8,5
NH ₄ ⁺	8	0,2 à 4 à pH 8
NO ₃ ⁻	146	1 à 10
PO ₄ ⁻⁻⁻	0,01	0,01 à 0,2
DCO	996	8

* Source : McNeely et coll., 1980 (Environnement Canada)

N.B. : Analyses faites en triplicate

lorsqu'on choisit ceux-ci, il est possible de tenir compte des niveaux écologiques considérés (producteurs, micro ou macro-consommateurs, transformateurs) et du genre d'informations recherchées (toxicité létale, sublétale ou chronique, bio-accumulation ou biodégradabilité de contaminants, etc.).

Lors du choix des essais biologiques à partir des différents facteurs énoncés, il faut adopter une démarche cohérente afin de pouvoir en intégrer les résultats. Pour une telle démarche, nous suggérons une évaluation basée sur trois niveaux de protection environnementale (Van Coillie et coll., 1984). Ces niveaux sont brièvement décrits au tableau 5.

L'évolution de l'écotoxicité entre ces trois niveaux de protection environnementale peut être représentée sous forme d'une spirale s'ouvrant progressivement à chaque niveau, comme le montre la figure 1.

Sur celle-ci, les trois niveaux de protection environnementale sont distincts en fonction de deux axes. Le premier axe sépare les effets létaux et sublétaux, tandis que le second différencie les effets directs ayant des manifestations toxiques aiguës à subaiguës, des effets insidieux menant à des manifestations toxiques chroniques. Ces répartitions concernent chacun des cinq niveaux trophiques des écosystèmes aquatiques.

L'application de cette conceptualisation pour évaluer l'écotoxicité du lixiviat des sédiments marins considérés est brièvement exposée au tableau 6.

TABLEAU 5

TROIS NIVEAUX DE PROTECTION ENVIRONNEMENTALE POUR L'ÉCOTOXICITÉ AQUATIQUE

NIVEAUX CONSIDÉRÉS	DÉRÈGLEMENTS		IMPACTS ÉCOTOXIQUES	
	CAUSES	CONSÉQUENCES	DIMENSIONS	PORTÉES
Premier	Pollution excessive (contaminants, agents pathogènes, conditions physico-chimiques altérées, etc.)	Disparition quasi-totale des composantes biotiques du milieu à l'exception d'espèces très tolérantes (certaines bactéries, par exemple)	Toxicité létale (aiguë à subaiguë)	Locale
Deuxième	Pollution sublétale causant un déséquilibre dans le milieu aquatique; ce type de pollution s'avère non bioaccumulable;	Déséquilibre de la diversité biologique et stress physiologique à court ou moyen terme	Toxicité sublétale (aiguë à subaiguë) résultant d'effets directs à court et moyen terme facilement mesurables	Régionale
Troisième	Toxicité insidieuse, parfois difficilement interprétable quant aux dangers réels qu'elle représente; elle n'est guère assimilable par le milieu récepteur et s'exerce par des mécanismes de bioaccumulation et/ou mutagénicité;	Réduction de la survie à long terme (taux de croissance et de reproduction des espèces), déstabilisation des structures et fonctions des écosystèmes aquatiques avec possibilité de récurrence d'effets létaux, induction de problèmes génotoxiques	Toxicité chronique résultant d'effets insidieux à long terme relativement difficiles à mesurer	Globale

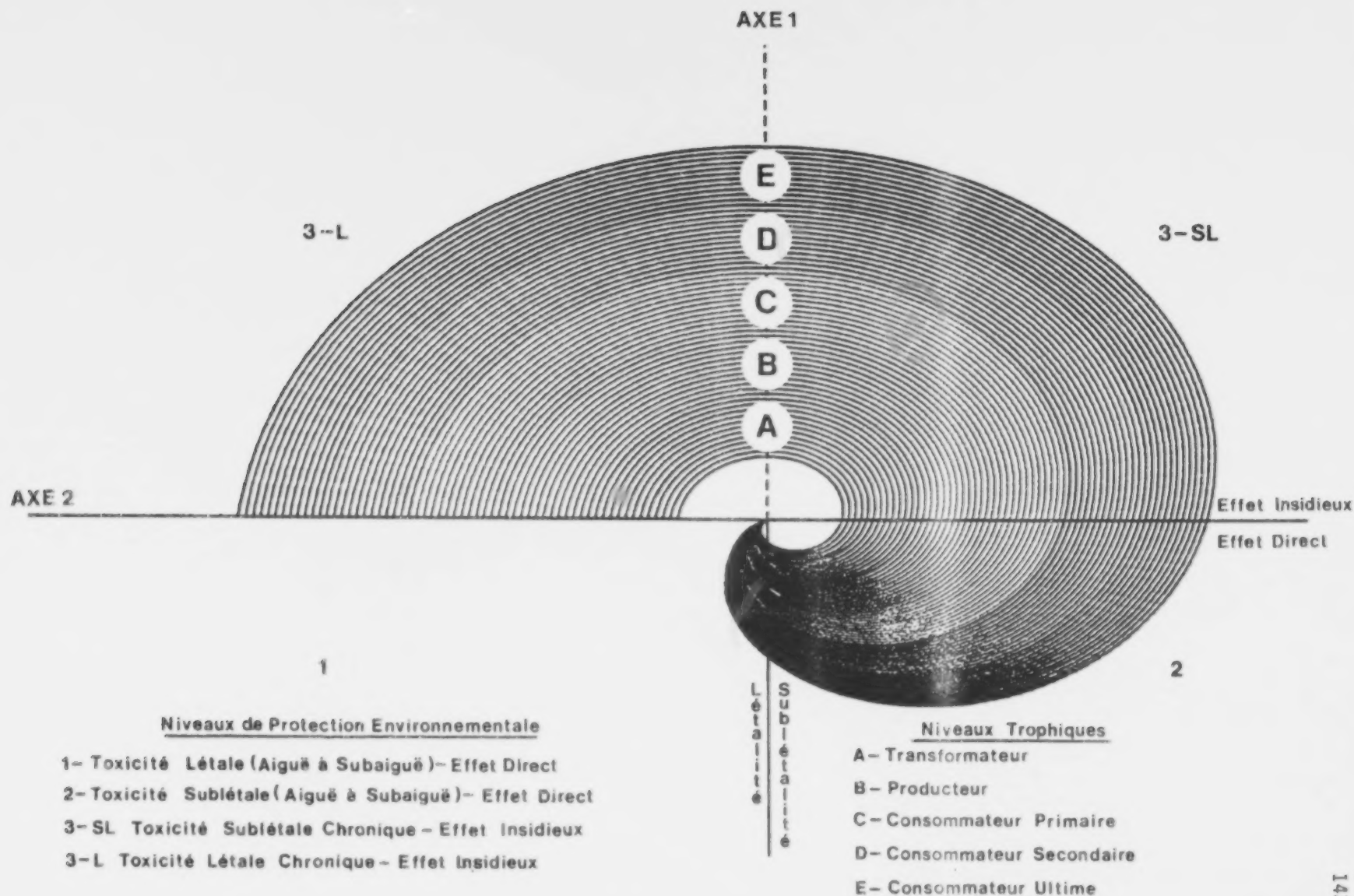


FIGURE I SPIRALE DE L'ECOTOXICITE .

TABLEAU 6
PROGRAMME DES ESSAIS BIOLOGIQUES EFFECTUÉS POUR
MESURER L'ÉCOTOXICITÉ DU LIXIVIAT DES SÉDIMENTS TESTÉS

NIVEAUX DE PROTECTION ENVIRONNEMENTALE	ESSAIS BIOLOGIQUES CHOISIS	RÉFÉRENCES MÉTHODOLOGIQUES	AVANTAGES	CONTRAINTES
PREMIER (TOXICITÉ LÉTALE)	Des tests de létalité avec des poissons marins n'ayant pu être réalisés pour des motifs de coût et de logistique; on a opté pour des essais biologiques avec des moules <u>Mytilus edulis</u> : leur survie a été observée durant 15 jours;	Montage : Spotte, 1979 Moules de la myti-culture Sambro près d'Halifax CL ₅₀ * : Stefan, 1977	Simplicité Représentativité Coût raisonnable	Sensibilité faible Longue durée "Reproductibilité" moyenne

* CL 50 : Concentration létale pour 50% des individus pendant une durée expérimentale précisée pour chaque test

TABLEAU 6
PROGRAMME ESSAIS BIOLOGIQUES EFFECTUÉS POUR
MESURER L'ÉCOTOXICITÉ DU LIXIVIAT DES SÉDIMENTS TESTÉS
(suite)

NIVEAUX DE PROTECTION ENVIRONNEMENTALE	ESSAIS BIOLOGIQUES CHOISIS	RÉFÉRENCES MÉTHODOLOGIQUES	AVANTAGES	CONTRAINTES
SECOND (TOXICITÉ SUBLÉTALE)				
a) Transformateur	Bioluminescence de <u>Photobacterim</u> <u>phosphoreum</u> (5 minutes à 15° C)	Beckman Instruments Inc., 1980 CI ₅₀ ** : Stefan, 1977	Rapidité Simplicité "Reproductibilité" Coût faible	Représentativité discutable Sensibilité moyenne
b) Producteur	Croissance des diatomées marines <u>Thalassiosira</u> <u>pseudonana</u> (8 jours à 18° C)	Milieu d'Harrison et coll., 1980 Sabatini, 1982 Souche axénique des laboratoires Bigelow CI ₅₀ ** : Stefan, 1977	Simplicité Sensibilité Représentativité	Longue durée Coût moyen à élevé "Reproductibilité" moyenne
c) Consommateur	Mobilité des larves (nauphii II ou III) des microcrustacés <u>Artemia salina</u> (24 heures à 25° C)	Leonhard, 1981 Van Haecke et Persoone, 1981 Souche de la compa- gnie Sanders CI ₅₀ ** : Stefan, 1977	Rapidité Simplicité "Reproductibilité" Coût faible	Représentativité discutable Sensibilité moyenne

** CI 50 : Concentration inhibant à 50% un paramètre physiologique pendant une durée expérimentale précisée pour chaque test

TABLEAU 6
PROGRAMME DES ESSAIS BIOLOGIQUES EFFECTUÉS POUR
MESURER L'ÉCOTOXICITÉ DU LIXIVIAT DES SÉDIMENTS TESTÉS
(suite)

NIVEAUX DE PROTECTION ENVIRONNEMENTALE	ESSAIS BIOLOGIQUES CHOISIS	RÉFÉRENCES MÉTHODOLOGIQUES	AVANTAGES	CONTRAINTES
TROISIÈME (TOXICITÉ CHRONIQUE)				
a) Transformateur	Mutagénicité chez <u>Salmonella typhimurium</u> pour la dépendance à l'histidine (3 jours à 35° C)	Ames et coll., 1975 Environnement Canada, 1984 Souches de types TA-97 et TA-100	Rapidité Simplicité "Reproductibilité"	Représentativité discutable Coût moyen à élevé Sensibilité moyenne à élevée
b) Producteur	Incorporation de la toxicité dans les diatomées marines <u>Thalassiosira Pseudonana</u> par mesure directe (toxicité du lysat cellulaire avant et après exposition) et par mesure indirecte (toxicité du milieu avant et après exposition) (4 heures à 18° C)	Blaise et coll., 1982 1982 Environnement Canada, 1984 Souche axénique des laboratoires Bigelow	Rapidité Simplicité Sensibilité Représentativité	Coût moyen à élevé "Reproductibilité" moyenne

TABLEAU 6
PROGRAMME DES ESSAIS BIOLOGIQUES EFFECTUÉS POUR
MESURER L'ÉCOTOXICITÉ DU LIXIVIAT DES SÉDIMENTS TESTÉS
(suite)

NIVEAUX DE PROTECTION ENVIRONNEMENTALE	ESSAIS BIOLOGIQUES CHOISIS	RÉFÉRENCES MÉTHODOLOGIQUES	AVANTAGES	CONSTRAINTES
TROISIÈME (suite) c) Consommateur	Bioaccumulation de contaminants dans les tissus mous et durs des moules <u>Mytilus edulis</u> (7 jours à 10° C)	Montage : Spotte, 1979 Bioaccumulation : McGreer et coll., 1981 Moules de la myti- culture Sambro près d'Halifax Analyses chimiques : voir paragraphe 3	Sensibilité Représentativité Simplicité rela- tive	Longue durée Coût élevé "Reproductibilité" moyenne à faible

Les résultats obtenus avec les six essais biologiques effectués en vue d'une évaluation écotoxicologique intégrée du lixiviat des sédiments du port d'Halifax sont synthétisés au tableau 7.

L'examen des résultats écotoxiques indique que les eaux de lixiviation des sédiments portuaires d'Halifax :

- i) n'exerçaient aucune écotoxicité létale détectable à court et moyen terme;
- ii) occasionnaient une écotoxicité sublétale très faible;
- iii) avaient un potentiel d'écotoxicité chronique, qui se traduisait par une mutagénicité et par un début de bioaccumulation de métaux lourds.

La portée de ces résultats, bien qu'utile pour une étude de la pollution du port d'Halifax, doit être envisagée dans le contexte méthodologique du présent rapport, ce qui fait l'objet du paragraphe suivant.

6. DISCUSSION

D'une part, les sédiments analysés contenaient divers contaminants reconnus toxiques et plusieurs de ceux-ci se retrouvaient dans l'eau de lixiviation.

D'autre part, le lixiviat des sédiments s'avérait très peu écotoxique aux niveaux létal et sublétal tout en ayant un potentiel de toxicité chronique qu'on décela au moyen de tests de mutagénicité et d'essais biologiques de bioaccumulation de métaux lourds.

TABEAU 7
ÉCOTOXICITÉ CONSTATÉE DU LIXIVIAT DES SÉDIMENTS TESTÉS

NIVEAUX DE PROTECTION ENVIRONNEMENTALE	ESSAIS BIOLOGIQUES EFFECTUÉS	RÉSULTATS ÉCOTOXIQUES		
		CI 50 *	AUTRES	UT **
PREMIER (TOXICITÉ LÉTALE)	<u>Mytilus edulis</u> : survie	100% : pas de mortalité	--	1
SECOND (TOXICITÉ SUBLÉTALE)				
a) Transformateur	<u>Photobacterium phosphoreum</u> : bioluminescence	100% : pas d'inhi- bition	--	1
b) Producteur	<u>Thalassiosira pseudonana</u> : . croissance . croissance après décompte de la stimulation de croissance imputable aux NO ₃ ⁻ et NH ₄ ⁺ des sédiments dans le lixiviat (voir tableau 4) ***	stimulation 50%	--	2

* CI 50 : Concentration inhibant à 50% un paramètre physiologique pendant une durée précise

** UT : Unité toxique = 100% : CI 50

*** Le décompte à effectuer est déterminé à partir du calcul de la fertilité des ions nutritifs NO₃⁻, NH₄⁺ et PO₄⁻³ du milieu pour la croissance de cultures végétales unicellulaires (Couture et coll., 1982; Van Coillie et coll., 1983b)

TABLEAU 7
ÉCOTOXICITÉ CONSTATÉE DU LIXIVIAT DES SÉDIMENTS TESTÉS
(suite)

NIVEAUX DE PROTECTION ENVIRONNEMENTALE	ESSAIS BIOLOGIQUES EFFECTUÉS	RÉSULTATS ÉCOTOXIQUES		
		CI 50 *	AUTRES	UT **
SECOND (suite)				
c) Consommateur	<u>Artemia salina</u> : mobilité	100% : pas d'in- hibition	—	1
TROISIÈME (TOXICITÉ CHRONIQUE)				
a) Transformateur	<u>Salmonella</u> <u>typhimurium</u> : mutagénicité	—	Concentration active minimale : 10%	10
b) Producteur	<u>Thalassiosira</u> <u>pseudonana</u> : incorporation de la toxicité ambiante (mesure directe et indirecte)	--	Facteur de bioac- cumulation de la toxicité **** : 1	1

**** Facteur de bioaccumulation de la toxicité :

- a) mesure directe : (UT du lysat cellulaire après exposition - UT du lysat cellulaire avant exposition) : UT du milieu avant exposition
b) mesure indirecte : (UT du milieu après exposition - UT du milieu avant exposition) : UT contrôle
(Blaise et coll., 1982; Van Coillie et coll., 1983a)

TABLEAU 7
ÉCOTOXICITÉ CONSTATÉE DU LIXIVIAT DES SÉDIMENTS TESTÉS
(suite)

NIVEAUX DE PROTECTION ENVIRONNEMENTALE	ESSAIS BIOLOGIQUES EFFECTUÉS	RÉSULTATS ÉCOTOXIQUES		
		CI 50 *	AUTRES	UT **
TROISIÈME (suite)				
c) Consommateur	<u>Mytilus edulis</u> : incorporation de métaux lourds et organochlorés	--	Facteurs de bioac- cumulation des con- minants***** dans les : tissus tissus mous durs Cd 7 1 Cr 1 1 Cu 1 30 Pb 18 1 Zn 10 1 BPC 1 DDT 1 HCB 1	

***** Facteur de bioaccumulation d'un contaminant :

[(Concentration du contaminant après exposition dans le tissu - concentration du contaminant avant exposition dans le tissu) - bioaccumulation éventuelle chez les témoins] : (Concentration du contaminant dans le milieu - concentration contrôle)
(MacLaren Marex, 1980; McGreer et coll., 1981)

Comment expliquer cette absence de toxicité aiguë et sub-aiguë du lixiviat de sédiments pourtant contaminés?

Quatre hypothèses peuvent être avancées.

- i) Les sédiments choisis étaient-ils insuffisamment contaminés?

Ceci semble plausible lorsqu'on considère les deux données suivantes : létalité pour le poisson Fundulus heteroclitus après 1 jour en présence des sédiments du port de Baltimore, E-U. (Tsai et coll., 1979) et mortalité élevée de la crevette Crangon septemspinosa après 2 à 4 jours dans un lixiviat de sédiments d'Inlet Albertini, Colombie-Britannique (EVS, 1977). Il convient toutefois de mentionner ici que la recherche de méthodes pour tester la toxicité des sédiments marins ne doit pas être restreinte à ceux qui sont extrêmement contaminés, mais doit plutôt s'appliquer à une large gamme de sédiments.

- ii) La texture essentiellement limoneuse des sédiments testés et leur matière organique (voir tableau 3) favorisaient-elles une rétention de leurs contaminants?

On peut le penser, car c'est un phénomène connu (Levings, 1983). Il faut cependant tenir compte de ce facteur dans la recherche méthodologique d'essais biologiques de toxicité applicables à divers types de sédiments, d'autant plus que les zones portuaires sujettes aux dragages renferment souvent des sédiments argilo-limoneux organiques.

- iii) La lixiviation faite était-elle trop peu agressive pour libérer les contaminants des sédiments en concentrations suffisamment toxiques?

C'est possible. Toutefois, amplifier le procédé de lixiviation en augmentant la durée ou la proportion de sédiments par rapport à l'eau ne semble pas devoir donner un meilleur rendement. En effet, selon Côté et Constable (1982), l'équilibre des contaminants entre les sédiments et le lixiviat est atteint après quelques heures et des rapports sédiments-eau voisins de 1:4 favorisent une stratification. Certes, en adoptant des pH acides ou basiques, la libération des contaminants associés aux sédiments marins est activée comme dans le cas des éluviations (Levings, 1983), mais ceci se révèle non représentatif pour des essais biologiques de toxicité et pour des conditions marines.

- iv) Les essais biologiques effectués pour déceler la toxicité létale et sublétale s'avèraient-ils insuffisamment sensibles et/ou trop courts?

Cette hypothèse apparaît vraisemblable à postiori, mais rappelons que l'objectif de la présente étude était une recherche de méthodes simples et rapides pour tester la toxicité de sédiments marins à draguer. Certes, il existe divers essais biologiques sensibles pour déceler la toxicité, mais ils se révèlent généralement coûteux et/ou longs, comme l'illustrent les exemples suivants : consommation d'oxygène (O'Connor et coll., 1977), évitement (Wildish et coll., 1977), bioaccumulation de contaminants durant 1 à 6 mois chez des vers annélides Nereis virens ou des palourdes Macoma balthica (Elder et coll., 1979; McLeese et coll., 1980; McGreer et coll., 1981, 1983 et 1984). De plus en plus, on vise plutôt à obtenir des essais biologiques non seulement sensibles, mais aussi relativement courts et économiques pour préciser l'écotoxicité des sédiments marins.

C'est dans cette tendance que se situent certaines études actuelles dont celle de Chapman (1984), préconisant des tests de toxicité létale de 10 jours avec l'amphipode Rhepoxinus abronius et des inhibitions de croissance des larves d'huîtres pendant 48 heures, ainsi que la nôtre proposant des tests de 3 à 7 jours pour déceler le potentiel de toxicité chronique.

7. REMARQUES

Afin de ne point alourdir l'exposé, nous avons préféré rassembler ici certaines données complémentaires.

i) Approche par diffusion

Tel qu'annoncé (voir paragraphe 2), cette approche a été expérimentée avec des suspensions sédiments-eau 1:4 à 1:20. Elle s'est cependant révélée peu efficace. En effet, après 8 jours de diffusion des sédiments du port d'Halifax en circuit fermé, on ne décela qu'une toxicité marginale (1,6 unité toxique) de ceux-ci (proportion 1:4) vis-à-vis des diatomées Thalassiosira pseudonana.

ii) Sédiments-témoins

Les analyses chimiques, la lixiviation, la diffusion et les essais biologiques ont été faits non seulement avec des fonds du port d'Halifax, mais aussi avec des sédiments marins pris en août 1983 dans un lieu non sujet à des pollutions dues à des contaminants, soit Baie des Sables (Québec). Ces sédiments-témoins étaient surtout sablonneux (60% de sable moyen et grossier < 0,25 µm et 35% de sable fin

de 0,06 à 0,25 μm) et ne renfermaient aucune concentration anormale de contaminants; leur lixiviat et leur diffusion n'occasionnaient aucune toxicité.

iii) Autres sédiments analysés

Des échantillons de sédiment furent également pris en août 1983 aux ports de Sydney (Nouvelle-Écosse) et de Baie-Comeau (Québec). Au premier lieu, ils ont été pris à un point situé à équidistance entre la "Pointe Battery" et l'extrémité sud de la jetée portuaire tandis qu'au second site, ils ont été prélevés de façon aléatoire et ensuite mélangés. Les sédiments du port de Sydney, de nature majoritairement limono-argileuse (77%), étaient tous contaminés par des métaux lourds (respectivement 6, 138, 199, 465 et 524 mg/kg en Cd, Cr, Cu, Zn et Pb), des cyanures (79 mg/kg) et des huiles (850 mg/kg). Par contre, les sédiments portuaires de Baie-Comeau, de texture essentiellement sablonneuse (95%) se révélaient moins contaminés (90 mg/kg pour les huiles et 1,8 mg/kg pour les BPC). On n'a pas pu, toutefois, compléter l'évaluation écotoxicologique de ces deux sédiments au moyen du programme d'essais biologiques après leur lixiviation ou diffusion.

iv) Bioaccumulation de toxicité versus bioaccumulation de contaminants

La première s'est avérée négative, alors que la seconde a montré qu'il y avait incorporation de métaux lourds à partir du lixiviat des sédiments du port d'Halifax (voir tableau 7). Ceci s'explique de la façon suivante : la première a été testée avec des diatomées (Thalassiosira pseudonana) après 4 heures d'exposition, laquelle durée ne

se révèle guère suffisante pour une bioaccumulation de métaux lourds dans les algues et, par extension, dans les diatomées, mais permettrait par contre une incorporation de contaminants organiques (Van Coillie et coll., 1983a et 1984) pour autant que ceux-ci soient présents en quantités élevées dans le milieu, ce qui n'est pas le cas pour le lixiviat considéré. Par ailleurs, la seconde bioaccumulation a eu lieu durant 7 jours dans des moules Mytilus edulis. Cette période a seulement permis de détecter le début du phénomène (facteurs de bioaccumulation 30); des études antérieures ont en effet montré que ce dernier, bien que présent après 7 jours, ne se stabilise de façon manifeste (facteurs de bioaccumulation 100) qu'après des durées plus longues (1 à 6 mois) chez les moules et les palourdes (EPS, 1980; McGreer et coll., 1981 et 1983).

v) Test de biodégradabilité

Afin de vérifier si la toxicité pouvait être masquée par de la matière organique et si elle était susceptible de se manifester ultérieurement lors d'une biodégradation de cette matière organique dans le lixiviat des sédiments du port d'Halifax, un test de biodégradabilité de ce lixiviat a été effectué. Il a duré 7 jours en conditions aérobies, selon une technique mise au point antérieurement (Van Coillie et coll., 1983a et 1984). Après ce test, la toxicité vis-à-vis de Photobacterium phosphoreum restait nulle et la DCO (demande chimique en oxygène) demeurait élevée (environ 1000 mg O₂/l). Ceci indique que la matière organique s'avérait peu biodégradable dans le lixiviat et que la toxicité de celui-ci n'était pas amplifiée par une biodégradation.

8. CONCLUSION

Notre étude montre que le choix d'essais biologiques simples et rapides pour tester l'écotoxicité de sédiments marins à draguer pour l'eau où ils seront rejetés doit tenir compte de la biodisponibilité réduite de leurs contaminants.

Toutefois, après lixiviation, leur potentiel d'écotoxicité chronique est décelable au moyen d'essais biologiques relativement simples et rapides, tels que des tests de mutagénicité ou de débuts de bioaccumulation durant 8 jours.

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BIOASSAY RESPONSES TO POLLUTANT CONCENTRATIONS
IN LAKE ST-LOUIS SEDIMENTS

P. E. Ross and H. Sloterdijk

Abstract

Lake St-Louis, formed by the confluence of the St-Lawrence and Ottawa rivers near Montreal, was the object of our sampling programme in 1984. Sediments were collected at 39 stations and analysed for physical characteristics, organic and inorganic pollutants, and microbial populations. Elutriates of these sediments were designated for chemical analyses and used as the contaminant solution in a suite of bioassays. These tests included the Microtox procedure (*Photobacterium*), an algal radiocarbon uptake assay (*Selenastrum capricornutum*), a rotifer mortality study (*Brachionus calyciflorus*), a series of cladoceran bioassays (*Simocephalus vetulus*, *Daphnia pulex*), and a nematode test (*Panagrellus redivivus*).

Among the 39 stations, metal concentrations were generally well-correlated with each other, but not with As, a metalloid. Organics were correlated with each other and with certain metals. Pollutant concentrations exceeded Ontario Ministry of Environment guidelines for open-water dredge spoil disposal at many stations. A composite ranking by degree of pollution is presented. The elutriation process resulted in significant liberation of Mn (32 stations), Fe (19 st.), Zn (14 st.), Cu (11 st.), Ni (1 st.) and Hg (1 st.). In most cases, sediment and elutriate concentrations for individual contaminants were significantly correlated.

The algal bioassay yielded the highest number of toxic responses, followed by the Microtox procedure and the nematode test. Only one concentration of elutriate (10%) was used in the nematode test, so this test could prove to be more sensitive when applied over the range of concentrations used in the other bioassays.

P.E. Ross and H. Sloterdijk

RESUME

Le lac St-Louis, formé par la confluence du Saint-Laurent et de la rivière Ontaouais près de Montréal, a été l'objet de notre programme d'échantillonnage en 1984. Des sédiments ont été collectés dans 39 stations et analysés pour leurs propriétés physiques, leurs polluants organiques et inorganiques, et les populations microbiennes. Les extraits de ces sédiments obtenus par lavage ont été désignés pour des analyses chimiques et utilisés comme solution contaminante dans une suite de tests biologiques. Ces tests incluaient la procédure Microtox (*Photobacterium*), un essai sur l'absorption du carbone radioactif par les algues (*Selenastrum capricornutum*), une étude de la mortalité des rotifères (*Brachionus calyciflorus*), une série d'essais biologiques sur les cladocères (*Simocephalus vetulus* *Daphnia pulex*) et un test sur les nématodes (*Panagrellus redivivus*).

Parmi les 39 stations, les concentrations en métaux possédaient en général une bonne corrélation avec celles des autres stations, sauf pour ce qui est de l'As, un métalloïde. Il y avait une corrélation des polluants organiques entre-eux de même qu'entre ces polluants et certains métaux. Les concentrations en polluants excédaient les lignes directrices du Ministère Ontarien de l'Environnement pour le dépôt, en "eau-ouverte", des résidus de drague et cela pour plusieurs stations.

Un composé classifié selon le degré de pollution est présenté. Le processus de séparation par lavage a résulté dans une libération considérable de Mn (32 stations), Fe (14 st.), Cu (11 st.), Ni (1 st.), et Hg (1 st.). Dans la plupart des cas, les concentrations issues des lavages et des sédiments pour les substances contaminantes individuelles avaient une corrélation significative.

Les tests biologiques sur les algues produisirent le nombre le plus élevé de réactions toxiques, suivis par la procédure Microtox et le test des nématodes. Seulement une concentration obtenue par lavage (10%) a été utilisée dans le test sur les nématodes, et c'est pourquoi ce test peut être plus sensible lorsque employé parmi l'éventail des concentrations utilisées pour les autres tests biologiques.

THE EFFECTS OF CHEMICAL TREATMENTS ON THE RETENTION AND
REDOX REACTIONS OF SELENIUM BY SELECTED FRESHWATER SEDIMENTS

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Selenite and selenate at submicrogram levels were added to suspensions of two lake sediments (Buffalo Pound and Katepwa Lakes) from the Qu'Appelle River Basin in southeastern Saskatchewan. The sediments were treated with either sodium hypochlorite for the removal of organic matter or with an acetylacetone-benzene solution for the removal of amorphous Al and Fe components. The treatment with sodium hypochlorite resulted in the cessation of the oxidation of selenite to selenate and the elimination of reduction of selenate to selenite by the Katepwa Lake sediment. The treatment also dramatically decreased the retention of selenite by the sediments, especially Katepwa Lake sediment. The retention of selenate by the sediments was not observed before or after the treatment. Upon the treatment with an acetylacetone-benzene solution, the Buffalo Pound Lake sediment lost the ability to oxidize selenite to selenate, and the retention of selenite by the sediment was dramatically decreased. This indicates that the sesquioxidic components were responsible for much of the sorptive capacity and also the oxidative abilities of the Buffalo Pound Lake sediment. The acetylacetone-benzene treatment did not affect the retention of selenite by the Katepwa Lake sediment as severely as by the Buffalo Pound Lake sediment, but the Katepwa Lake sediment lost the ability to reduce selenate to selenite. This was attributed to the inhibition of biotic activity and the associated biochemical processes by the acetylacetone-benzene treatment. These results indicate that these two sediments are very different in nature, a result of the differing physical conditions in the lakes.

Keywords: selenium, sediments, pretreatments, minerals, organics, oxidation, reduction, adsorption.

Les effets des traitements chimiques sur la
rétention et les réactions d'oxydoréduction du
sélénium par des sédiments d'eau douce sélectionnés.

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RESUME

Du sélénite et du sélénate, à un niveau inférieur au microgramme ont été additionnés aux sédiments en suspension de deux lacs (Buffalo Pound et Katepwa) à partir du bassin de la rivière Qu'Appelle au sud-est de la Saskatchewan. Les sédiments ont été traités avec soit; du sodium hypochlorite, pour l'enlèvement des matières organiques ou soit avec une solution de benzène-acétylacétone pour l'enlèvement des composants amorphes de Al et de Fe. Le traitement avec le sodium hypochlorite a résulté en la cessation de l'oxydation du sélénite en sélénate et l'élimination de la réduction du sélénate en sélénite par les sédiments du lac Katepwa. Le traitement a aussi décrû dramatiquement la rétention du sélénite par les sédiments, spécialement ceux du lac Katepwa. La rétention du sélénate n'a pas été observée avant ou après le traitement. Durant le traitement avec la solution au benzène-acétylacétone, les sédiments du lac Buffalo Pound perdirent la capacité d'oxyder le sélénite en sélénate, et la rétention du sélénite par les sédiments a été dramatiquement décrûe. Ceci indique que les composants "sesquioxydiques" ont été responsables, pour une grande part, de la capacité d'absorption et aussi de la capacité d'oxydation des sédiments du lac Buffalo Pound. Le traitement au benzène-acétylacétone n'a pas affecté la rétention du sélénite

par les sédiments du lac Kaptewa aussi sévèrement que les sédiments du lac Buffalo Pound; mais les sédiments du lac Katepwa ont perdu la capacité de réduire le sélénate en sélénite. Ceci a été attribué à l'inhibition de l'activité biotique associée avec les processus biochimiques du traitement au benzène-acétylacétone. Ces résultats indiquent que ces deux sédiments sont de par nature très différents; un résultat de la différenciation dans la condition physique des lacs.

INTRODUCTION

Selenium is a trace element whose paucity understates its importance. It is an essential nutrient for animals, including human beings, and its role in the prevention of a wide range of diseases is under study. Metabolized selenium is antagonistic to Hg, As and Cd, decreasing the toxicity of these metals to aquatic and terrestrial organisms. Selenium is also a toxic element with a small concentration difference between essential and toxic quantities which underlines the significance of the processes which govern its bioavailability. For this reason an understanding of natural processes and the impact of man's activities on the availability of selenium is essential in order to avoid the contamination or depletion of selenium in natural environments.

Huang et al. (1983) reported that sediments from Katepwa Lake in Qu'Appelle River basin in Saskatchewan were capable of reducing selenate to selenite, while the sediment from Buffalo Point Lake in the basin performed the opposite reaction. Lipinski (1985) subsequently found that the oxidation of selenite to selenate was an abiotic reaction, and postulated that the reduction of selenate to selenite was mediated by biotic activity, associated biochemical processes, and/or organic matter.

The objective of this study was to investigate the effects of chemical treatments of the sediments on their reactions with selenite and selenate and the significance in understanding the role of sediment components in selenium transformations in freshwater systems.

MATERIALS AND METHODS

SEDIMENTS

The lake sediments studied were from Buffalo Pound and Katepwa Lakes in southeastern Saskatchewan. They are located in the Qu'Appelle Valley, a geographical feature which resulted from melt-water erosion in the last ice age. The nature of these lakes and sediments are described elsewhere (Oscarson et al. 1981). Bottom sediment samples were collected with an Ekman dredge. Several sediment samples from each lake were combined and thoroughly mixed to make a composite sample which was used for all experiments. The sediment samples were stored in sealed plastic bottles at 4°C.

REAGENTS

Reagent grade Na_2SeO_3 and Na_2SeO_4 were standardized gravimetrically (Erdey 1965). The procedure given by Wilkie and Young (1970) was followed for the preparation of the 2,3-diaminonaphthalene (DAN) complexing reagent.

CHEMICAL TREATMENTS OF SEDIMENTS

Sodium Hypochlorite Treatment. The sediments were treated with sodium hypochlorite (Anderson 1963) to remove organic matter from the sediment samples. A 20 mL aliquot of 4-6% sodium hypochlorite solution freshly adjusted to pH 9.5 was added to a 10 g sediment sample in a 100 mL centrifuge tube. The tube was placed in a boiling water bath

for 15 min, and then centrifuged for 5-10 min at 1400g. The supernatant was decanted and analyzed for the extracted elements by plasma arc spectrometry (the analysis was done by the Saskatchewan Research Council - Analytical Services Dept.). The treatment was repeated three times. The sediment was then placed in dialysis tubes to remove the remaining sodium hypochlorite. At the end of the treatment the sediment was resuspended in water and stored at 4°C in a glass bottle. The pH was adjusted with dilute acetic acid to approach that of untreated sediments.

Acetylacetone-Benzene Treatment. A 100 mL aliquot of 5% acetylacetone in benzene solution was added to 10 g of air-dried sediment. The suspension was kept at room temperature with occasional shaking for a period of 200 hours. At the end of this period the sediment suspensions were centrifuged for 5-10 min at 1400 g and the supernatant was decanted and analyzed for the elements extracted using the same procedure which was employed for the analysis of the supernatants from the sodium hypochlorite treatment. The sediment was washed in diethyl-ether. Final washing was carried out in water to remove the last traces of the organic solvents. The sediment was stored at 4°C. The pH was adjusted by addition of dilute acetic acid to approach that of untreated sediments.

REDOX REACTIONS OF SELENITE AND SELENATE

The chemically treated composite sediment samples from Buffalo Pound and Katepwa Lakes were used for this study. Sediment samples were suspended in deionized-distilled water at a ratio of 1 g (oven-dry weight basis) of sediment to 100 mL of solution which contained 100 ng Se as selenite or selenate per mL of solution.

For all experiments, the flasks containing the suspensions were stoppered and placed on a Blue M oscillating shaker at a speed of 80-90 oscillations·min⁻¹ in a water bath at 4, 25 or 60°C. At various time periods up to a maximum of 7 weeks, the suspensions were thoroughly mixed and an aliquot was withdrawn. The suspension was centrifuged at 1400g for 30 min and the supernatant was filtered using cellulose nitrate membrane filters (0.45 µm pore size). The filtrate was analyzed fluorometrically in duplicate for selenite and selenate by the method of Huang et al. (1983) as modified by Lipinski (1985).

At each sampling period, the E_h (measured with a Pt electrode vs. a AgCl/Ag reference electrode in 4 M KCl) and pH of the suspensions were determined.

RESULTS AND DISCUSSION

SODIUM HYPOCHLORITE TREATMENT

The treatment of sediments with sodium hypochlorite was given as an alternate method to the hydrogen peroxide treatment for the removal of organic matter (Jackson 1979). When selenate was added to the untreated Katepwa Lake sediment there was a rapid reduction of selenate to selenite, depleting selenium from solution (Fig. 1). When the sediment was treated with sodium hypochlorite, the observed reduction and adsorption reactions ceased. The reduction of selenate and the

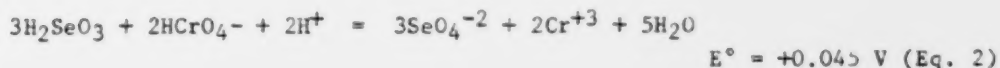
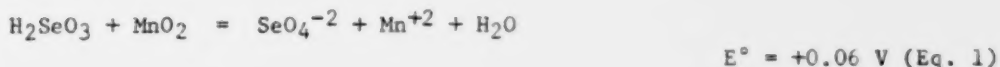
subsequent retention of selenium by the Buffalo Pound Lake sediment was not observed either before or after treatment with sodium hypochlorite (Fig. 1).

The oxidation of selenite to selenate occurred when selenite was added to the untreated sediments (Fig. 2). Compared with Katepwa Lake sediment, the reaction proceeded more rapidly in the case of Buffalo Pound Lake sediment. After the sodium hypochlorite treatment of both sediments, no oxidation reactions were observed.

In both the reduction of selenate by Katepwa Lake sediment and the oxidation of selenite by both sediments, the reactions were halted after treatment with sodium hypochlorite. This observation indicates two possibilities: (1) the materials which caused the oxidation or reduction were removed by the treatment of the sediments, and (2) the treatment while not removing the active material(s) rendered them ineffective in their interactions with selenate or selenite. The data indicate that the reduction of selenate to selenite was a reaction mediated by living microorganisms and/or by the byproducts of microbial activity. The removal or inactivation of the organic sediment components and/or biota by the sodium hypochlorite treatment apparently caused the inhibition of the reduction process.

The elemental analysis of the supernatant of the sodium hypochlorite treated sediments (Table 1) indicates that significant amounts of materials other than organic matter were extracted by the treatment. The kinds of inorganic materials removed by this treatment seem to fall under two categories. The first includes elements associated with organic matter and the second, inorganic complexes loosely associated with the mineral components of the sediments. These inorganic complexes may include sesquioxides and other hydrous oxides which may be present as discrete colloidal precipitates or as coatings on surfaces of clay minerals. They may also exist as complexes with a series of inorganic ligands, low molecular weight organic acids, and functional groups of humic substances of sediments (Huang & Kozak 1970; Schnitzer & Kodama 1977; Schwertmann & Taylor 1977; Kwong & Huang 1978; Huang 1980). These materials are part of the most active segment of soils and sediments. The removal of this fraction of the sediments may explain the cessation of the oxidation of selenite to selenate by the sediments.

Of the elements removed by the sodium hypochlorite treatment (Table 1), only two are thermodynamically feasible to oxidize selenite to selenate:



The mediation of the conversion of selenite to selenate by oxygen is also feasible ($E^\circ = 0.079 \text{ V}$). Another possibility is cobalt,

but the reaction is thermodynamically favorable only in 3M HNO₃ ($E^{\circ} = 0.31$ V).

Although the oxidation of selenite to selenate by manganese dioxide is thermodynamically feasible, this reaction was not observed after 7 wks (Huang et al. 1983). The oxidation of selenite to selenate did not occur spontaneously in deionized-distilled water. The oxidation of selenite to selenate which is mediated by manganese dioxide or oxygen, although thermodynamically favorable, appears to be very slow.

Adsorption of selenite was also affected by the sodium hypochlorite treatment of the sediments (Table 2). Before the treatment, the Katepwa Lake sediment had a higher sorption capacity than did the Buffalo Pound Lake sediment. After the treatment, no oxidation of selenite to selenate occurred and the Buffalo Pound Lake sediment retained more selenite than did the Katepwa Lake sediment (Fig. 2). This phenomenon is attributable to the removal of organic components and certain reactive hydrous oxides of the sediments. Buffalo Pound Lake sediment had a lower organic carbon content but had a finer texture than the Katepwa Lake sediment (Oscarson et al. 1981). After the sediment was treated with sodium hypochlorite the retention of selenite by the remaining fraction of the sediment was less severely affected, apparently owing to its finer texture. Furthermore, the elimination of the oxidation of selenite to selenate by the Buffalo Pound Lake sediment after the sodium hypochlorite treatment evidently counteracted the decrease of the adsorption of selenite by the removal of reactive components of the sediment.

Sesquioxides and their complexes with silica are widely distributed in both the colloidal and non-colloidal fractions of the Katepwa and Buffalo Pound Lake sediments (Oscarson et al. 1981). These sesquioxidic components could play an important role in governing the dynamics of selenium in these aquatic systems. Carbonates of calcium and magnesium are also present in large quantities in these sediments. These carbonates may armor sediment oxide surfaces, thereby reducing their sorptive capacity and decreasing reaction rates (Oscarson et al. 1981).

Coarse particles as well as the finer particles have significant sorption capacity for solutes (Hwang et al. 1976; Gibbs 1977; Huang & Liaw 1978). This can be attributed, at least in part, to surface coatings of hydrous aluminum, iron and manganese oxides on crystalline minerals and complexation of these oxides with humic substances (Jenne 1968; Huang 1980). Therefore, the removal of these sesquioxidic and humic components by the sodium hypochlorite treatment very drastically decreased the adsorption of selenite by the sediments.

ACETYLACETONE-BENZENE TREATMENT

Acetylacetone, when added to soil as a 5% solution in benzene specifically removes oxides and hydroxides of aluminum and iron with efficiency similar to the pyrophosphate treatment, with minimal impact on the organic component of the sediment (Giovannini & Sequi 1976). Table 3 shows that this treatment did in fact remove large quantities of aluminum and iron while removing generally lesser amounts of the other elements than did the sodium hypochlorite treatment.

The adsorption of selenite by both sediments was affected by the acetylacetone-benzene treatment (Table 2). Compared with the untreated sediment the adsorption of selenite by the Katepwa Lake sediment after the acetylacetone-benzene treatment was very significantly decreased (Fig. 3), but was higher than that which occurs after the sodium hypochlorite treatment (Fig. 2). Compared with the sodium hypochlorite treated sediment (Fig. 2), the Buffalo Pound Lake sediment lost more of its retention capacity for selenite after the acetylacetone-benzene treatment (Fig. 4), indicating the importance of hydrous oxides and hydroxides of aluminum and iron in the retention of selenite. This substantiates the observation of previous workers (Olson 1939; Plotnikov 1964; Hingston et al. 1967; Hamdy & Gissel-Nielsen 1977). Ryden et al. (1977) studied the mechanisms of phosphate adsorption by hydrous ferric oxide gels. In this case the phosphate becomes part of the structural unit of the ferric oxide. This reaction may also be possible with selenite, and may explain both the high sorption capacity of ferric hydroxides for selenite and tenacity with which selenite is held by ferric compounds. When the sediments were treated with acetylacetone-benzene, hydrous oxides of aluminum and iron were preferentially removed. These sesquioxidic components were probably present as coatings on the mineral surfaces of the sediments and complexed with organic matter.

Rajan and Watkinson (1976) postulated that on allophane clays selenite exchanged with hydroxyl, adsorbed sulfate and adsorbed silicate. The hydroxyl groups released probably existed as edge hydroxyls (M-OH), rather than structural, bridging hydroxyl groups (M-OH-M). Selenite with its trihedron structure is an unsuitable structural substitute, whereas phosphate groups may replace structural silicate (Rajan & Watkinson 1976). Selenite may also exchange with aquo groups.

Rajan (1979) gave three possible reaction mechanisms for the retention of selenite by hydrous alumina. The first reaction neutralizes the positive charge on the alumina, releasing an aquo group to solution as the selenite is retained. The second releases an hydroxyl into solution. The last mechanism releases an aquo molecule to solution and leaves a negatively charged surface. It is important to note that selenite is adsorbed as HSeO_3^- , displacing one ligand on the hydrous alumina. Unlike phosphate groups, which evidently are able to break the aluminum polymers, the adsorption of selenite reaches a maximum when the adsorbent surface becomes neutral in charge (Rajan et al. 1974; Rajan 1975; 1976).

The retention of selenite by Katepwa Lake sediment was less severely affected by the acetylacetone-benzene treatment (Fig. 3) than by the treatment with sodium hypochlorite (Fig. 2). The acetylacetone-benzene treatment removed similar amounts of iron and aluminum from both sediments (Table 3). The nature of sesquioxidic components in the Katepwa Lake sediments may differ from the Buffalo Pound Lake sediments and other sediment components could also be responsible for some of the observed retention of selenite by the Katepwa Lake sediment.

When selenate was added to the acetylacetone-benzene treated Katepwa Lake sediment there was no discernible reduction of selenate to selenite (Fig. 5). Apparently the acetylacetone-benzene treatment had inhibited biotic activity and/or altered the organic fraction to render it ineffective in the reduction of selenate to selenite. This alteration may have removed certain metals (Table 1) which serve as inorganic coenzymes. Selenate was not added to the treated Buffalo Pound Lake sediment since there were no detectable interactions between selenate and the untreated Buffalo Pound Lake sediment.

When selenite was added to the acetylacetone-benzene treated Katepwa Lake sediment (Fig. 3) the reaction was similar to that observed when selenite was added to untreated Katepwa Lake sediment at 60°C (not shown). Initially a rapid oxidation of selenite to selenate appeared to occur. After three days, however, the selenate levels in solution had decreased and remained at a low level for the remaining reaction period. Likewise, the selenite levels in solution fell rapidly initially, but the selenite level in solution increased after 3 d and remained relatively constant for the rest of the reaction period. This indicates that a complex reaction was occurring in the acetylacetone-benzene treated or heated (60°C) sediments. It may be that the reactive agents which were activated by heating the untreated Katepwa Lake sediment were similarly affected by the acetylacetone-benzene treatment.

When selenite was added to the Buffalo Pound Lake sediment which had been treated with acetylacetone-benzene, the oxidation reaction of selenite was also suspended (Fig. 4). This indicates that the components removed by the acetylacetone-benzene treatment are vital in mediating the oxidation of selenite.

These two sediments from the Buffalo Pound and Katepwa Lakes were very different with respect to their interactions with selenite and selenate, despite similar basic mineralogy, climate, and anthropogenic and natural inputs. Buffalo Pound is a shallow lake (3.0 m mean depth) which is aerated for most of the year, while Katepwa Lake is deep (14.4 m mean depth) and the sediments undergo anaerobiosis for much of the year. The differences in physical characteristics of the lakes have modified the chemical and biological properties of the sediments which has in turn affected the interactions of these sediments with selenium.

SUMMARY AND CONCLUSIONS

The effects of chemical treatment on the oxidation of selenite and reduction of selenate and the sorption of these selenium species by freshwater sediments from Katepwa and Buffalo Pound Lakes in the Qu'Appelle River basin in Saskatchewan, Canada were studied.

When the two sediments were treated with sodium hypochlorite, all redox reactions of selenate and selenite were halted. This suspension in the reduction of selenate to selenite by the Katepwa Lake sediment after the treatment was attributed to the elimination of biotic processes and/or the removal of organic components. The sodium hypochlorite treatment also reduced the ability of the two sediments to

retain selenite, indicating the importance of the sediment fraction removed by the treatment in the retention of selenite.

Removal of the sesquioxidic components of the sediments by the acetylacetone-benzene treatment suspended the ability of the Buffalo Pound Lake sediment to oxidize selenite to selenate and substantially reduced the retention of selenite by the sediment. This indicates the importance of the sesquioxidic fraction of the sediment in the retention and oxidation of selenite by the Buffalo Pound Lake sediment. The adsorption of selenite by the Katepwa Lake sediment was also reduced by the acetylacetone-benzene treatment, but not as severely as when treated with sodium hypochlorite, a result of a substantially higher organic matter content in the Katepwa Lake sediment. There are no interactions between selenate and acetylacetone-benzene treated Katepwa Lake sediment.

ACKNOWLEDGEMENTS

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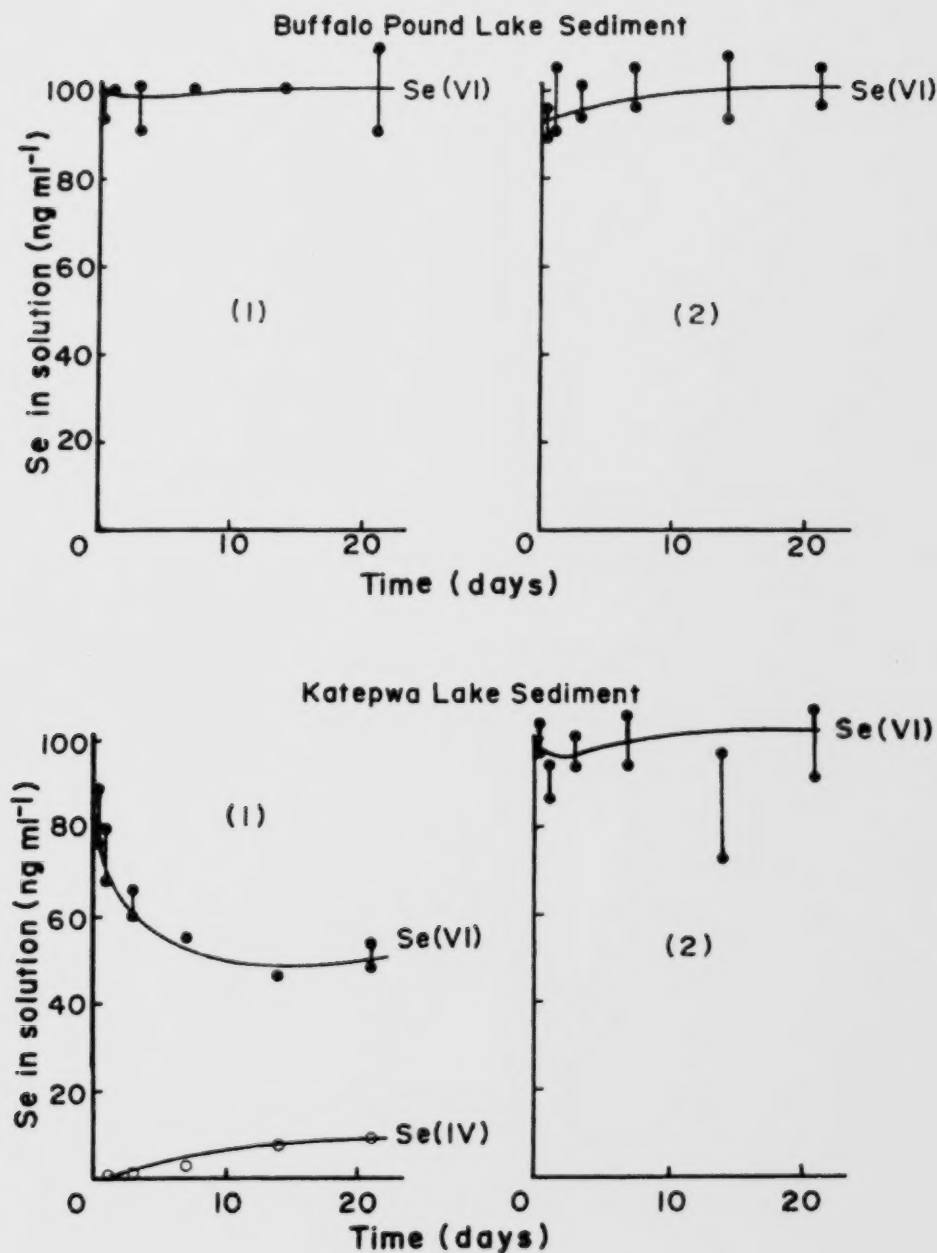


Figure 1. The reduction of selenate to selenite at 25°C by Buffalo Pound and Katepwa Lake sediments as influenced by the sodium hypochlorite treatment: (1) Before Treatment and (2) After Treatment.

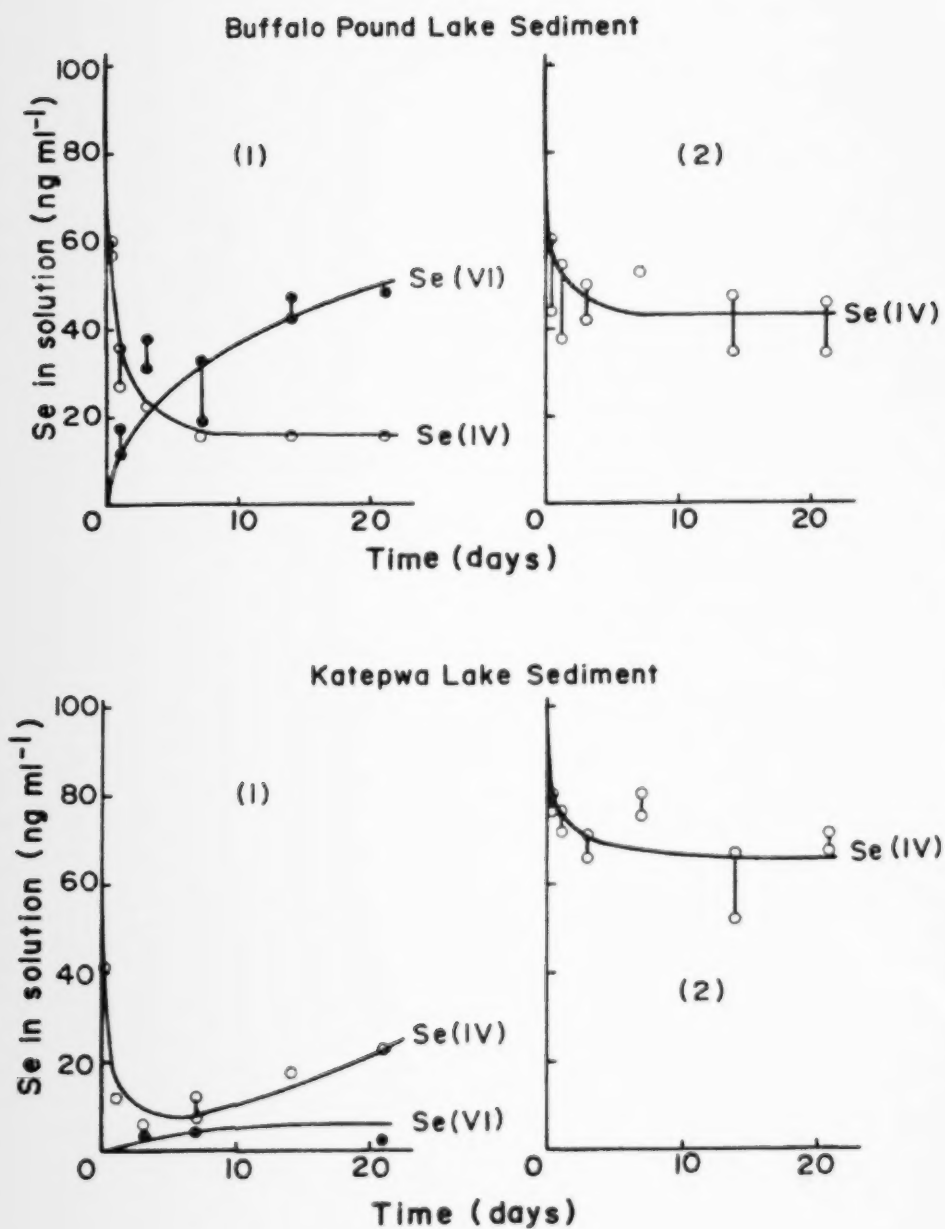


Figure 2. The oxidation of selenite to selenate at 25°C by Buffalo Pound and Katepwa Lake sediments as influenced by the sodium hypochlorite treatment: (1) Before Treatment and (2) After Treatment.

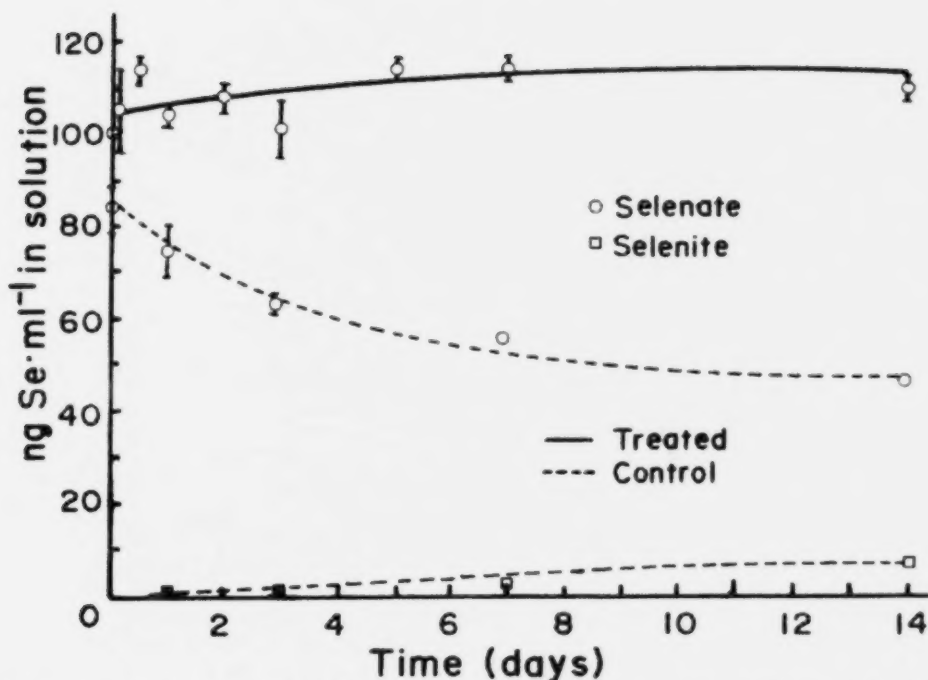


Figure 3. Interactions of selenite with Katepwa Lake sediment at 25°C after the acetylacetone-benzene treatment. If no deviation is indicated, the standard deviation is within the size of the symbol.

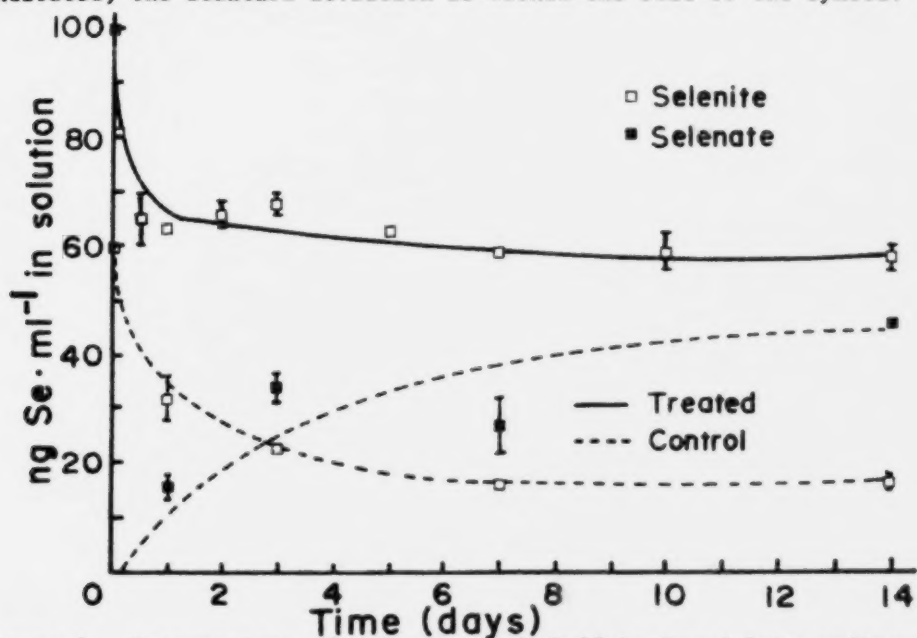


Figure 4. Interactions of selenite with Buffalo Pound Lake sediment at 25°C after the acetylacetone-benzene treatment. If no deviation is indicated, the standard deviation is within the size of the symbol.

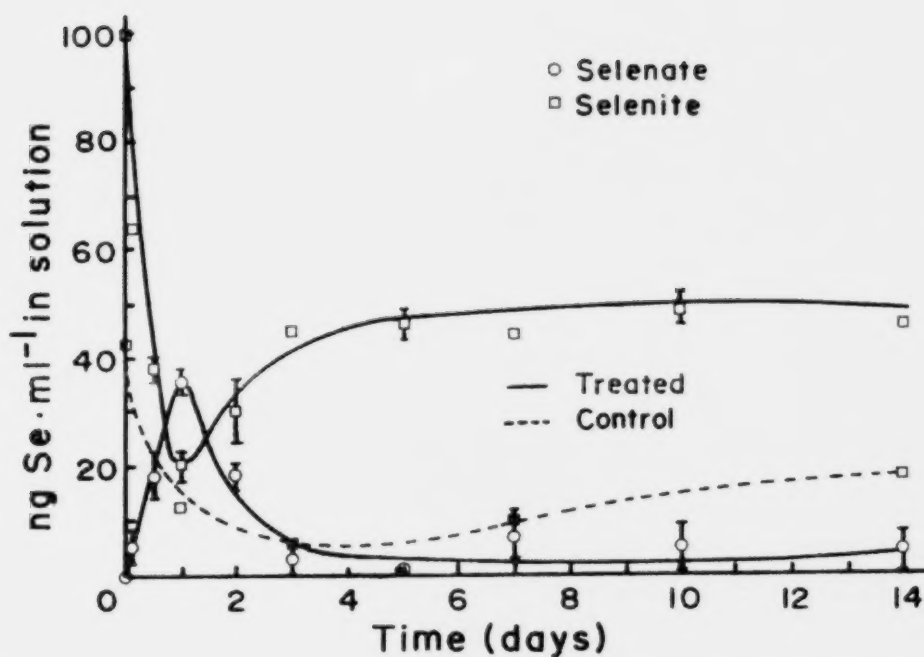


Figure 5. Interaction of selenate with Katepwa Lake sediment at 25°C after the acetylacetone-benzene treatment. If no deviation is indicated, the standard deviation is within the size of the symbol.

Table 1. Elemental analysis of the supernatant of the NaOCl treated Katepwa and Buffalo Pound Lake sediments.

Element	mmol of elements released . kg ⁻¹ of sediment	
	Buffalo Pound Lake sediment	Katepwa Lake sediment
Ca	84.2	137.2
Mg	50.4	65.8
K	19.8	25.6
Si	5.61	14.1
P	2.82	5.57
B	1.18	1.85
Al	0.16	0.70
Fe	0.06	0.34
Mn	0.60	2.23
Ba	0.26	0.17
Zn	0.14	0.17
Co	0.01	0.01
Cr	0.04	0.06
Cu	0.06	0.12
Ni	0.05	0.10
Ti	n.d.*	0.01
Pb	0.01	n.d.
Mo	n.d.	0.07

* not detectable; Be, Cd, V, W and Ag were not detectable

Table 2. Retention of selenite by Buffalo Pound and Katepwa Lake sediments at the end of a 7-day reaction period at 25°C, after treatment with either sodium hypochlorite or acetylacetone in benzene.

Treatment	Final pH	Final Eh (mV)	ng Se(IV) .mL ⁻¹ in solution*	ng Se(VI) .mL ⁻¹ in solution*	µg Se(IV) retained.g ⁻¹ of sediment
<u>Buffalo Pound Lake sediment</u>					
Control	8.0	400	24 ± 1.7	23 ± 3.6	5.4 ± 0.4
NaOCl	7.5	420	40 ± 4.7	nd**	6.0 ± 0.5
Ace-Ben ^α	7.6	430	57 ± 4.6	nd	4.3 ± 0.5
<u>Katepwa Lake sediment</u>					
Control	7.7	430	9 ± 0.8	2 ± 0.0	9.1 ± 0.1
NaOCl	7.8	430	69 ± 7.7	nd	3.1 ± 0.8
Ace-Ben ^α	7.3	450	32 ± 3.1	6 ± 3.1	6.2 ± 0.3

* The initial Se concentration in solution was 100 ng Se(VI) . mL⁻¹
and 0 ng Se(IV) . mL⁻¹

** Not detectable

α Acetylacetone in benzene

Table 3. Elemental analysis of the supernatant of the acetylacetone in benzene treated Katepwa and Buffalo Pound Lake sediments.

Element	mmol of elements released . kg ⁻¹ of sediment	
	Buffalo Pound Lake sediment	Katepwa Lake sediment
Ca	21.8	32.3
Mg	19.5	38.5
K	5.45	9.77
Na	12.2	55.7
P	2.52	3.13
B	1.79	3.46
Al	5.00	3.52
Fe	17.6	21.6
Mn	2.04	0.67
Ba	0.06	0.07
Zn	0.10	0.16
Co	0.01	0.01
Cr	0.01	0.01
Cu	0.05	0.02
Ni	0.03	0.02
Mo	n.d.*	0.04
Be	0.03	0.04
V	0.04	0.04

* not detectable; Ti, Pb, Cd, W and Ag were not detectable

FISH-WATER-SEDIMENT RELATIONSHIPS IN A
MERCURY CONTAMINATED WATERCOURSE

J.W. Parks, J.A. Sutton, C. Currie, N. Sims, J.D. Hollinger

Mercury concentrations in biota, waters, and sediments of the Wabigoon-English River system remain markedly elevated, although major loadings of inorganic mercury from a now defunct chlor-alkali plant were sharply reduced in 1970. To develop relationships between mercury levels in and between various biotic and abiotic compartments, we standardized sampling procedures and sites from field studies conducted on the Wabigoon-English River system between 1978 and 1980.

For the biotic compartment, we collected young-of-the-year and yearling northern pike (*Esox lucius*), yearling yellow perch (*Perca flavescens*) and crayfish (*Orconectes virilis*) annually. These biota were selected because: 1) they were allopatric, 2) they lived in a very localized area; 3) they belonged to different trophic levels, and 4) they usually had life spans for which exposure data were available or could be estimated. For the water compartment, total and methylmercury concentrations were averaged from mean monthly data for the August 1978 - May 1980 period. Surface sediments (0-5 cm) were also collected for total mercury analysis. The sediments were qualitatively similar, comprised of clay and silt overlain by a loose brown organic floc.

Mean mercury concentrations in biota, waters, and sediments were log transformed and correlation coefficients derived between young of the year and yearling pike; yearling perch; crayfish; methylmercury and total mercury in water; and, total mercury in sediment. All 21 correlations were statistically significant at the 0.05 level.

Despite differences in habitat, diet, uptake routes and proportions of methylmercury bodyburden, a strong correlation ($r=0.98$, $p<0.001$, $n=8$) between mercury in pike and mercury in crayfish was observed. Similar, highly significant relationships were observed for total mercury-methylmercury in water; total mercury in sediments-methylmercury in waters; and, total mercury in sediments-total mercury in waters. The results of these field studies suggest that laboratory investigations of the cycling of diffuse source contaminants (i.e. sediments) could be very fruitful.

J.W. Parks, J.A. Sutton, C. Currie, K. Suns, J.D. Hollinger

RESUME

Les concentrations en mercure dans les biota, l'eau et les sédiments du système des rivières Wabigon-English restent remarquablement élevées même si des déversements majeurs de mercure inorganique, d'un plant de chlore-alkali maintenant inopérant, ont été diminués drastiquement en 1970. Pour établir des relations entre les niveaux de mercure à l'intérieur et entre différents compartiments biotiques et abiotiques, nous avons standardisé les procédures d'échantillonnage et de location provenant des études sur le terrain conduites dans le système des rivières Wabigon-English entre 1978 et 1980.

Pour les parties biotiques, nous avons collecté annuellement; des brochets du nord (*Esox lucius*) nés de l'année et d'autres ayant un an d'âge, des perches jaunes (*Percas flavescens*) et des écrevisses (*Orconectes virilis*) ayant un an d'âge. Ces biotas ont été choisis parce que: 1-ils vivaient dans différentes locations, 2-ils vivaient dans un espace spécifique, 3-ils appartiennent à différents niveaux trophiques, 4-ils avaient habituellement une longévité pour laquelle les données d'exposition étaient disponibles ou pouvaient être estimées. Une moyenne a été faite pour les concentrations totales et de "méthylemercure" des compartiments d'eau à partir des données moyennes mensuelles pour la période d'août 1978 à mai 1980. Les sédiments de surface (0-5 cm) ont aussi été collectés pour l'analyse complète du taux de mercure. Les sédiments étaient qualitativement similaires, comprenant de la glaise et du sable fin recouvert par une mousse organique brune.

Les concentrations moyennes en mercure dans les biota, l'eau et les sédiments ont été transformées en logarithme et les coefficients des corrélations, dérivés entre les brochets jeunes de l'année et ceux âgés d'un an; les perches âgées d'un an; les écrevisses; le "méthyle mercure" et le niveau total de mercure dans l'eau; et le niveau total de mercure dans les sédiments. Les 21 corrélations étaient toutes statistiquement significatives à un niveau de 0.05.

En dépit des différences dans l'habitat, la diète, les routes d'absorption et les proportions d'accumulation corporelle en "méthyle mercure", une forte corrélation ($r=0.98$ $P < 0.001$, $n = 8$), entre le mercure chez le brochet et celui chez l'écrevisse, a été observée. Une relation similaire, hautement significative a été observée pour le total de mercure - "méthyle mercure" dans l'eau; le total de mercure dans les sédiments - "méthyle mercure" dans l'eau; et le total de mercure dans les sédiments - le total de mercure dans l'eau. Les résultats de ces études sur le terrain suggèrent que les investigations de laboratoires sur le cycle de diffusion des sources de contaminants (i.e. sédiments) peut s'avérer être utiles.

Abstract: EFFECTS OF LOW AMBIENT PH ON GILL MORPHOLOGY OF
FRESHWATER FISH

by

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P7B 5E1

Gill morphology was examined in white suckers (Catostomus commersoni) kept in near-neutral and acidified freshwater media using light microscopy as well as scanning and transmission electron microscopy. Gills collected from fish in near-neutral pH media had chloride cells (CC) residing mainly in the interlamellar regions of filament epithelia and along the bases of lamellar epithelia while mucous cells (MC) predominated along the edges of filament epithelia. Gills, either exposed to ambient pH = 4 for 12 - 96 h or collected from an acid lake at pH = 5.3, showed no deterioration in structure but rather had changes in the relative distribution and abundance of both CC and MC, particularly on lamellar epithelia. However, there was no marked alteration in either the apical surface morphology, cytoplasmic contents or size of either cell type in acid-exposed gills.

It is proposed that the proliferation of gill MC in acid-exposed suckers may have served as a protective barrier both to prevent epithelial damage and to reduce diffusional ion loss. The gill CC proliferation probably occurred to enhance active ion influx. These findings will be discussed in relation to those available in the literature on other fish species similarly exposed to external acid stress.

Helve Høbe

RESUME

La morphologie des branchies a été examinée chez les "suceurs blancs" (*Cotostomus commersoni*) gardés dans des médium d'eau douce s'approchant de la neutralité en pH et d'un autre ayant été acidifié. L'observation a été faite en utilisant une micrographie lumineuse de même qu'avec celle de diffusion et transmission d'électrons. Les branchies, obtenues des poissons placés dans un médium dont le pH s'approchait de 7, avaient des cellules chloriques (CC) principalement logées dans les régions inter-lamellaires des filaments épithéliaux et le long des parties inférieures des lamelles épithéliales alors que les cellules muqueuses (CM) prédominaient le long des côtés des filaments épithéliaux. Les branchies exposées à un pH ambiant de 4 durant 12 à 96 hr, ou soit obtenues d'un lac acide avec un pH égale à 5.3, ne montraient aucune détérioration structurelle mais présentaient plutôt des changements dans la distribution relative et l'abondance des deux (CM) et (CC), particulièrement sur les lamelles épithéliales. Cependant, il n'y avait pas d'altérations marquées pour la morphologie apicale de surface, de même que pour la grosseur et le contenu cytoplasmique des deux types de cellules à l'intérieur des branchies exposées au milieu acide.

Il est proposé que la prolifération des (CM) branchiales chez les suceurs exposés à l'acide peut avoir servi de barrière protectrice pour prévenir des dommages épithéliaux et pour réduire la diffusion occasionnant une perte d'ions. La prolifération des (CC) branchiales survient probablement pour augmenter l'afflux d'ions actifs. Ces constatations vont être discutées en relation avec celles disponibles dans la littérature et traitant des autres espèces de poisson exposées de façon similaire à un stress acide externe.

THE USE OF TOXICOLOGICAL INFORMATION FOR
WATER QUALITY MANAGEMENT IN ONTARIO

Ontario's water quality management goal is to ensure that surface waters are of a quality which is satisfactory for aquatic life and recreation. By meeting these requirements other uses such as drinking water and agriculture will usually be protected.

To achieve the goal a series of criteria known as the Provincial Water Quality Objectives have been developed. Traditionally these criteria have been derived from an evaluation of short-term acute toxicity tests and, where available, sub-acute evaluations, tainting and bioaccumulation studies.

In order to expand or revise the list of criteria, we rely heavily on basic aquatic toxicological research. As one would expect, for traditionally accepted contaminants such as ammonia and some metals, substantial volumes of aquatic toxicity data exist and, surprisingly, there appear to be reasonable amounts of sound toxicological data for many of the more recent substances of concern such as the industrial organics chlorinated phenols and chlorinated benzenes.

However, as the science of detecting more and more substances at lower and lower concentrations evolves, the job of a water resource manager becomes more complex. The identification of "new" substances in a water or effluent sample creates the immediate demand for an aquatic protection criterion. To fulfill our role, we depend heavily upon the aquatic toxicologist to carry out the basic research.

To compound the issue, water resource managers must also consider a laundry list of other factors including: additive, synergistic and antagonistic effects of mixtures, partitioning, food chain dynamics, bioconcentration factors, half-lives, environmental fate ... the list goes on. While we cannot count on aquatic toxicologists to address all of these issues, we are surely looking at that community to help us unravel some of the complex problems.

What are we doing in Ontario to encourage new initiatives in aquatic research? Through in-house initiatives and funding of research at universities, for example, we are encouraging a range of studies.

At Lakehead University, for example, we are supporting a long-term study of the acute and chronic effects of individual industrial organic compounds on fish. As well as assessing the effects of individual substances the researchers here are evaluating the combined effects of two or more chemicals with the ultimate aim of developing a technique for mathematically modelling mixtures of contaminants.

Elsewhere, we are supporting research into the uptake of selected contaminants by aquatic organisms such as algae, invertebrates, and fish and then evaluating food chain dynamics using these organisms. We are also supporting research that will lead us to an understanding of the role of environmental contaminants related to fish tumours.

To assist in field studies, we are using young fish, adult fish, clams and invertebrates as bioaccumulators, caged fish to measure toxic effects. We intend through joint field/laboratory programs to establish correlations between what we can observe in the field and simulate in the laboratory. Again, we will count heavily on the research efforts of aquatic toxicologists in universities, government and private consulting to lead us in these efforts.

I began this presentation talking about the development of water quality protection criteria but evolved to a much broader range of issues. It is obvious that, to understand and effectively manage our aquatic environment, the resource manager, working in close cooperation with aquatic toxicologists and others, must understand the complex nature of that environment; make his needs known and encourage, through financial and moral support, the fundamental research required.

John G. Ralston, Manager
Aquatic Contaminants Section
Water Resources Branch
Ontario Ministry of the Environment
June 18, 1985

John G. Ralston

RESUME

Le but du contrôle de la qualité de l'eau en Ontario est d'assurer que l'eau de surface soit d'une qualité satisfaisante pour la vie aquatique et la récréation. En rencontrant ces exigences, d'autres usages comme pour l'eau potable et l'agriculture seront généralement protégés.

Pour atteindre ce but, une série de critères connus sous les Objectifs Provinciaux de la Qualité de l'Eau été développée. Traditionnellement ces critères ont été dérivés d'une évaluation des tests de toxicité sur la sensibilité à court terme, lorsque disponibles, des évaluations sous le niveau sensible, des études de contamination et d'accumulation biologique. Pour élargir ou réviser la liste des critères, nous dépendons largement des recherches de base sur la toxicologie aquatique. Comme on peut l'espérer pour des contaminants traditionnellement acceptés comme l'ammoniac et certains métaux, il existe un volume substantiel des données de toxicité aquatique et, surprenamment, il semble y avoir un nombre raisonnable de données complètes de toxicologie pour plusieurs nouvelles substances d'intérêt comme les phénols organiques industriels chlorés et les benzènes chlorés.

Quoi qu'il en soit, comme la science détecte un nombre de substances de plus en plus grand à des concentrations de plus en plus petites, le travail du contrôleur des ressources de l'eau devient plus complexe. L'identification des "nouvelles" substances dans un échantillon d'eau ou d'un effluent crée une demande immédiate pour des critères de protection aquatique. Pour remplir notre rôle, nous dépendons grandement sur les toxicologistes aquatiques pour mener à bien les recherches de base.

Pour regrouper les résultats, les contrôleurs des ressources de l'eau doivent aussi considérer une liste contenant d'autres facteurs comme; les additifs, les effets synergistes et antagonistes des mélanges le partitionnement, la dynamique de la chaîne alimentaire, les facteurs des concentrations biologiques, les déterminants de l'environnement ... et la liste continue. Bien que nous ne pouvons compter sur les toxicologistes aquatiques pour diriger leurs efforts, sur tous ces facteurs, nous regardons sûrement vers cette communauté pour nous aider à clarifier certains de ces complexes problèmes.

Que faisons-nous en Ontario pour encourager de nouvelles initiatives de recherches aquatiques. Au travers des initiatives de l'organisme et du financement de recherches dans les universités, par exemple, nous encourageons une vaste étendue d'études.

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Par exemple, à l'Université Lakehead, nous supportons une étude à long terme sur les effets sévères et chroniques des composés organiques individuels de l'industrie sur les poissons. De même que l'évaluation des effets individuels des substances, les chercheurs évaluent aussi les effets combinés de deux ou plusieurs produits chimiques avec l'intention ultime de développer une technique pour faire des modèles mathématiques sur les mélanges des contaminants.

Ailleurs, nous supportons la recherche sur l'absorption, de contaminants choisis, par des organismes aquatiques comme les algues, les invertébrés, les poissons et ensuite est fait l'analyse de la dynamique de la chaîne alimentaire incorporant ces organismes. Nous supportons aussi des recherches qui vont mener à la compréhension du rôle des contaminants sur l'environnement reliés aux tumeurs des poissons. Pour assister les études sur le terrain, nous utilisons de jeunes poissons, des poissons adultes, des moules et des invertébrés comme accumulateurs biologiques et des poissons encagés pour mesurer les effets toxiques. Nous voulons en joignant les programmes de laboratoires et ceux sur le terrain, établir des corrélations entre ce que nous pouvons observer sur le terrain et simuler en laboratoires. Ici encore, nous allons compter grandement sur les efforts de recherche des toxicologistes aquatiques des universités et sur les consultants privés et gouvernementaux pour nous diriger dans ces efforts. J'ai commencé cette présentation en parlant du développement des critères de protection pour la qualité de l'eau et j'ai évolué dans un contexte plus large touchant plusieurs secteurs. Il est clair que, pour comprendre et contrôler efficacement notre environnement aquatique, le directeur des ressources, travaillant en étroite collaboration avec les toxicologistes aquatiques et autres, doit comprendre la nature complexe de cet environnement; faire connaître ses besoins et encourager aux travers d'un support moral et financier, la recherche fondamentale requise.

Estimating Data for Risk Assessment Models

G. Veith

As our understanding of chemical exposure, toxicology, and ecosystem responses matures, it is natural for there to be a proliferation of models which attempt to integrate this knowledge into a useful package. Attempts to model risks have included the use of many dissimilar techniques varying from simulation models of ecosystems to a form of expert system based on heuristic models of our ecosystem response knowledge. Risk models may attempt to reflect important processes in ecosystems, or they may be policy models which reflect the mind of informed administrators. Whatever the scope and approach of risk assessment models, the models attempt to give decision makers a sense of the relative effects of different scenarios on a specific issue.

The underpinnings of all risk assessment models for toxic chemicals are the laboratory data used in the comparison with constructed risk criteria. The Toxic Substances Control Act and the use of discharge permits are causing the regulatory community to realize that the number of untested chemicals exceed those tested by 50 to 1. Consequently, elaborate risk assessment models will be of limited value unless the data requirements for the models evolve with the resources to produce data. One solution is the development of the technology to estimate the results of laboratory testing directly from chemical structure. This technology, called structure-activity relationships, is actually a complex series of models of the activity of chemical structures as they interact with water, suspended solids, and biological systems.

One of the interesting results of the use of structure-activity models with risk assessment models is that the risk criteria may be reformulated in terms of structural properties directly rather than be formulated in terms of biological hazard. Bioaccumulating chemical residues in ecosystems have put many populations at risk since 1940. Early work in structure-activity relationships showed that bioaccumulation of chemicals (at least in aquatic organisms) was dependent on a chemical property (n-octanol/water partition coefficient) and metabolism. In only a few years, the risks associated with bioaccumulation were estimated directly from the partition coefficient which can be computed in a few seconds rather than from a \$6,000 test.

This presentation will give one view of models for risk assessment and how they will interface with models of chemical systems.

G. Veith

RESUME

Alors que notre compréhension vis-a-vis des réponses à l'exposition aux produits chimiques, à la toxicologie et à l'écosystème se développe, il est naturel alors d'y avoir une prolifération des modèles essayant d'intégrer ce savoir dans un cadre pratique. Les essais pour incorporer les risques à ces modèles ont inclut l'usage de plusieurs techniques différentes variant d'un modèle de simulation de l'écosystème à un genre de système d'expert basé sur les réponses de l'écosystème. Les modèles de risques peuvent essayer de refléter des processus importants de l'écosystème, ou ils peuvent être des modèles politiques reflétant l'esprit d'administrateurs informés. Quel que soit l'étendue et l'approche de l'évaluation des modèles de risques, ils essaient de procurer à ceux qui prennent les décisions, un sens des effets relatifs aux différents scénarios d'un sujet spécifique.

Les fondations de tous les modèles d'évaluation des risques pour les produits chimiques sont les données de laboratoires utilisées en comparaison avec les critères de risques établis. L'Acte de Contrôle des Substances Toxiques et l'usage des permits de déversement forcent les groupes de régularisation à réaliser que le nombre de produits chimiques non-testés dépasse ceux testés par une relation de 50 pour 1. Conséquemment, les modèles élaborés d'évaluation des risques vont être d'une valeur limitée à moins que les exigences pour les données des modèles se développent avec les ressources pour produire les données. Une solution possible est le développement de la technologie pour estimer les résultats des tests de laboratoires directement des structures chimiques. Cette technologie appelée, relation de la structure et de l'activité, est actuellement une série complexe de modèles de l'activité des structures chimiques lorsqu'elles entrent en interaction avec l'eau, les solides en suspension et les systèmes biologiques.

Un des résultats intéressants quant à l'usage des modèles de la structure et de l'activité avec ceux de l'évaluation des risques est que les critères de risques peuvent être reformulés directement en termes de propriétés structurales plutôt que de l'être en termes de dangers biologiques. L'accumulation biologique des résidus chimiques dans l'écosystème a mis en danger plusieurs populations depuis 1940. Les premiers travaux sur la relation de la structure et de l'activité montraient que l'accumulation biologique des produits chimiques (du moins pour les organismes aquatiques) était dépendante d'une propriété chimique (n-octanol/coefficient de division de l'eau) et du métabolisme. En quelques années seulement, les risques associés avec l'accumulation biologique ont été estimés directement du coefficient de dispersion qui peut être calculé en quelques secondes au lieu d'utiliser un test coûtant \$6000.

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Cette présentation va donner un point de vue des modèles d'évaluation des risques et comment ils inter-agissent avec les modèles des systèmes chimiques.

ENVIRONMENTAL ASSESSMENT FOR A MINING PROJECT

Menno Speyer

Abstract

In 1983-84 Noranda Inc. conducted an environmental assessment for the new Hemlo Division Golden Giant Mine located near Marathon in North-west Ontario. This presentation reviews the environmental concerns that were resolved during the planning and pre-operational phase in the development of the mine. These include site selection for tailings disposal, water supply, effluent treatment and the discharge of treated effluent. Engineering studies of alternatives were conducted to assist tailings site and water supply selections. Special techniques were employed in the construction of the tailings dam to provide rigorous seepage control. Metallurgical pilot plant studies successfully delineated the gold process configuration and provided solutions allowing preliminary testwork for cyanide treatment alternatives that could be optimized under actual operating conditions. Environmental baseline studies conducted in the two affected watersheds will allow future comparisons to predicted impacts. Although the Ontario Environmental Assessment Act is not generally applicable to private undertakings, Noranda Inc. agreed with the Ministry of the Environment to present the plans to the local communities and media. Open houses and two public information sessions were held in the nearby communities to ensure that community concerns about the environment had been adequately addressed. Ministry of the Environment approvals for tailings disposal, effluent treatment and discharge were given only after full consideration of public input. Noranda's resolution of the aforementioned environmental aspects of the development through proper planning and engineering design is presented.

Menno Speyer

RESUME

En 1983-84 la compagnie Noranda Inc. effectuait une évaluation de l'environnement pour le nouveau plan de "Hemlo Division Golden Giant Mine" situé près de Marathon dans le Nord-Ouest de l'Ontario. Cette présentation revoie les préoccupations pour l'environnement qui ont été résolues durant la phase de planification et pré-opératoire du développement de la mine. Ceci inclu la sélection d'un site pour le dépôt des résidus de l'approvisionnement d'eau, le traitement des effluents et le déversement des effluents traités. Des études d'ingénieries ont été conduit pour différentes alternatives visant à appuyer le site de dépôt des résidus et la sélection de l'approvisionnement en eau. Des techniques spéciales ont été employé pour la construction du barrage pour les résidus dans le but de procurer un contrôle rigoureux des fuites. Les études d'un plan métallurgique pilote ont décrit avec succès la configuration du processus aurifère et procure des solutions permettant le travail préliminaire de tests sur les alternatives de traitement du cyanide qui pouvait être optimisé à l'intérieur des conditions opérationnelles actuelles. Les bases des études sur l'environnement conduites pour les deux réservoirs d'eau affectée vont permettre des comparaisons futures sur les prédictions des impacts. Même si l'Acte de l'Ontario pour l'Evaluation de l'Environnement n'est généralement pas applicable aux engagements privés, Noranda Inc., s'est mis d'accord avec le Ministère de l'Environnement pour présenter les plans aux communautés locales et aux médias. L'ouverture à tous du plan de même que deux sessions publiques d'information ont été tenu dans les communautés avoisinantes pour s'assurer que les préoccupations de ces communautés sur l'environnement aient été adéquatement satisfait. Les approbations du Ministère de l'Environnement pour le site de dépôt des résidus, le traitement et déversement des effluents ont été octroyé seulement après une pleine considération des apports publics. La résolution de Noranda sur les aspects précédemment mentionnés de l'environnement quant au développement au travers d'une planification adéquate et de conceptions d'ingénieries est présentée.

RESEARCH PRIORITIES

W.M.J. Strachan

1) THE CANADIAN COUNCIL OF RESOURCE AND ENVIRONMENT MINISTERS AT THEIR OCTOBER 1984 MEETING REQUESTED TWO THINGS OF THEIR TASK FORCE ON WATER QUALITY A) THE PRODUCTION OF CANADIAN WATER QUALITY GUIDELINES FOR 5 USES OF WATER

- i) AQUATIC LIFE AND WILDLIFE.
- ii) RAW WATER SUPPLY FOR DRINKING WATER.
- iii) RECREATIONAL WATER AND AESTHETIC VALUES
- iv) AGRICULTURE; LIVESTOCK
IRRIGATION.
- v) INDUSTRIAL.

AND B) A REPORT HIGHLIGHTING RESEARCH PRIORITIES TO PROVIDE INFORMATION TO ENABLE MORE GUIDELINES TO BE WRITTEN AND RECOMMENDATIONS ON HOW TO IMPLEMENT THE RESEARCH NEEDS.

2) ACTIVITIES TO DATE:

a) THE GUIDELINES ARE BEING WRITTEN BY A TEAM OF SCIENTISTS AND REVIEWED BY THE PROVINCES, TERRITORIES AND APPROPRIATE FEDERAL DEPARTMENTS. DUE TO TIME CONSTRAINTS, ONLY PUBLISHED REVIEWS WERE USED UNLESS AN IMPORTANT NEW PIECE OF INFORMATION WAS POINTED OUT TO THE PRODUCTION TEAM.

b) THE MATERIAL WAS STUDIED TO SEE WHETHER IT COULD BE APPLIED TO CANADIAN CONDITIONS, IF NOT, AN ATTEMPT WAS MADE TO MODIFY IT. THIS WAS NOT ALWAYS POSSIBLE.

c) WHILST DEVELOPING GUIDELINES FOR EACH USE, THE AUTHORS WILL HIGHLIGHT AREAS WHERE GUIDELINES CANNOT BE DEVELOPED OR ARE INADEQUATE BECAUSE MORE INFORMATION IS NEEDED. THESE NEEDS FOR INFORMATION WILL FORM THE RESEARCH PRIORITIES REPORT TO CCREM HANDOUTS WILL BE AVAILABLE AT THE BEGINNING THE WORKSHOP (NOV 5) GIVING DETAILS ON SOME OF THE INFORMATION GAPS WHICH HAVE BEEN IDENTIFIED..

d) ONCE THE FIRST EDITION OF THE GUIDELINES IS ACCEPTED BY CCREM, WORK WILL BEGIN ON UPDATING THEM USING DATA PUBLISHED SINCE THE DATE OF THE REVIEWS WHICH WERE USED TO OBTAIN THE INITIAL INFORMATION.

3) EMPHASIS IS SHIFTING FROM CHEMICAL ANALYSIS OF WATER, ALONE, TOWARDS AN UNDERSTANDING OF THE DYNAMICS OF THE AQUATIC ENVIRONMENT. CONSEQUENTLY, SEDIMENTS AND BIOTA ARE BECOMING MUCH MORE SIGNIFICANT IN THE OVERALL ASSESSMENT OF ENVIRONMENTAL QUALITY. FOR EXAMPLE, SEDIMENTS CAN PLAY AN IMPORTANT ROLE IN THE FATE OF TOXIC CHEMICALS, PARTICULARLY HEAVY METALS. AQUATIC ORGANISMS, IN THEIR CAPACITY AS BIOCONCENTRATORS, BIOMAGNIFIERS, AS WELL AS DISPLAYERS OF CHRONIC AND SUBCHRONIC EFFECTS, ARE A CHOICE OF MEDIUM WITH WHICH TO RELATE CHEMICAL ANALYSIS AND MORE ACCURATELY ASSESS AQUATIC CONDITIONS ("HEALTH" OF THE AQUATIC ENVIRONMENT). LEVELS IN DIFFERENT MEDIA SHOULD BE RELATED TO WATER CHEMISTRY IF POSSIBLE, AS BACKTRACKING TO INDUSTRIAL EFFLUENTS MAY BE NEEDED. THESE RECOMMENDATIONS COULD ALSO FORM

PART OF THE REPORT TO CCREM.

4) THE CANADIAN WATER QUALITY GUIDELINES ATTEMPT TO INDICATE WHICH CHARACTERISTICS OF WATER AFFECT THE TOXICITY OF CHEMICALS AND WHY THEY SHOULD BE TAKEN INTO ACCOUNT WHEN PROPOSING SITE SPECIFIC WATER QUALITY OBJECTIVES. WATER QUALITY DIFFERS WIDELY ACROSS CANADA AND THE GUIDELINES CAN ONLY PROVIDE A GENERAL DISCUSSION ON TOXIC EFFECTS AND THEN GIVE A RECOMMENDATION FOR EACH CHEMICAL. THIS THEN HAS TO BE MODIFIED TO SUIT THE LOCAL WATER TYPE. AT PRESENT, ONLY GENERAL COMMENTS CAN BE MADE, BUT IT IS HOPED THAT RESEARCH WILL GENERATE SOME ANSWERS OR FORMULAE WHICH WILL MAKE THE TASK OF CONSIDERING SUCH QUALITIES AS PH, HARDNESS, CHELATION ETC. MUCH EASIER. THESE NEEDS COULD BE IN THE CCREM RESEARCH PRIORITIES REPORT.

5) IN THE REPORT TO COUNCIL IN 1986 ON RESEARCH PRIORITIES FOR WRITING THE WATER QUALITY GUIDELINES, IT IS PLANNED TO ITEMISE AS MUCH OF THE NEEDED RESEARCH AS POSSIBLE, AND ALSO HOW TO IMPLEMENT IT. THIS WILL MEAN THAT FIELDS OF STUDY WILL BE DETERMINED AND PROPOSALS MADE FOR FUNDING. AREAS OF EXPERTISE WILL BE SOUGHT IN BOTH THE PRIVATE AND GOVERNMENT SECTORS.

THE OBJECTIVE OF THE "MINI-WORKSHOP" IS TO DISCUSS SOME OF THE RESEARCH GAPS WHICH HAVE COME OUT OF THE CCREM EXERCISE, AND POSSIBLY GIVE SOME INSIGHT INTO WHERE WE GO FROM HERE. THIS WILL AID IN WRITING THE REPORT TO THE CCREM AND MAY HELP IN ENSURING THAT THE SCARCE RESEARCH DOLLARS GIVE THE BEST RETURNS.

PROBLEMS IN DETERMINING THE WATER SOLUBILITY OF
ORGANIC COMPOUNDS

A. BHARATH, C. MALLARD, D. ORR, G. OZBURN and A. SMITH

ABSTRACT: The amount of a slightly soluble organic compound contained in a given volume of water is a function of how the solute was introduced into the aqueous phase and of how the aqueous phase was treated. This amount of solute may not represent the true solubility of the compound.

A. Bharath, C. Mallard, D. Orr,
G. Ozburn and A. Smith

RESUME

La quantité d'un composé organique légèrement soluble contenu dans un certain volume d'eau est fonction de la manière dont le soluté est introduit dans la phase aqueuse et de la manière dont celle-ci a été traitée. Cette quantité de soluté peut ne pas représenter la vraie solubilité du composé.

HEALTH CONSIDERATIONS FOR
BIOASSAY FISHES

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Health Considerations for Bioassay Fishes

Toxicity bioassays rely on the response of an animal to measure the effect of a specific toxicant or group of toxicants (Sprague, 1973). The measured affects are most often lethal (LC50, LD50), physiological (growth, fecundity) and behavioral (avoidance). All of these measurements, much like any system of measurement, rely heavily on the integrity of the measuring device. Yet when we use fish as the measuring device in bioassay systems, the fish are seldom tested to see if they are in working order before we ask them to measure a toxicological effect. Since pathological conditions in apparently healthy animals can significantly alter the animals ability to accurately measure the effect of a toxicant (Doull, 1980), it would be prudent to evaluate fish before they are used to measure the effects of toxicants.

This paper describes pathological lesions found in fish thought to be free from disease and describes some of the effects such lesions would have on the ability of the fish to measure the effects of toxicants. These cases are taken from the files of the Fish Pathology Laboratory, Ontario Veterinary College.

CASE I NITRITE TOXICITY

Yearling rainbow trout were obtained from a certified hatchery for use in an experiment in which changes in hematological values were going to be used to measure the affect of the experimental treatment. The control fish were held in the control system and the test fish were placed in a test system which had been used several times without problems. However, fish placed in the test system became lethargic and darkly pigmented within one week. A histopathological and clinical pathological examination revealed that these fish had slightly brown coloured gills, severe gill thrombosis

and aneurisms and a mild normocytic anemia. Because these pathological changes directly affected the hematological values in these fish, it would not be possible to separate out the effects of the toxicant from those associated with the pathologic changes.

In this case the pathologic changes are consistent with nitrite toxicity and the production of methemoglobinemia. Levels of nitrite in the water were found to be several times above levels recommended for the maintenance of healthy fish. This case demonstrates that although the fish were without infectious disease, the presence of environmentally induced pathology would have profoundly affect the results of the experiment. Any data gathered from this experiment would reflect the conditions of the experimental set up rather than the presence or absence of the toxicant.

CASE II AMOEBIC GILL DISEASE

Examinations for parasites at the time of certification are restricted to two protozoan parasites (Department of Fisheries and Oceans, 1984). This case describes another protozoan disease found in experimental fish which had been obtained from a certified source. The control fish had a normal appearance while the test fish grow poorly were lethargic, had increase opercular rates, increased coughing rates and mildly increase mortality. Histopathological examination revealed the presence of markedly hyperplastic gill epithelium and the presence of a protozoa on the surface of the gill. Electron microscopic examination of these parasites suggest that they were likely an amoebae (Daoust and Ferguson, 1985). In this case the entire response of the fish in the treatment group could be attributed to the disease in the fish rather than a response to the experimental manipulations.

CASE III CEREBRAL DIASTOMONIASIS

Fathead minnows (Pimephales promelas) were obtained from the wild for use in the evaluation of a variety of environmental conditions. Behavioral response was to be used as one of the chief unit of measurement. Some mortalities were occurring in the control lot and the investigators thought that the deaths should be diagnosed as a routine precaution prior to undertaking the experiments.

The fish had a normal appearance grossly. Histopathological examination revealed the presence of large metacercaria encysted within the brain case. The brain damage caused by the presence of these numerous metazoan parasites would likely alter the central nervous system functions and likely produce behavioral responses which reflect pathology of the nervous system rather than the experimental manipulation.

These cases serve to demonstrate the wide range of severe pathological conditions found in fish being used in research. Several other examples of pathological conditions seen in fish from certified hatcheries and/or experimental fish by the Fish Pathology Laboratory (1981 to 1985) are listed below:

VIRUS

Infectious Pancreatic Necrosis

BACTERIAL

Furunculosis
Bacterial Kidney Disease
Hemorrhagic Septicemia
Columnaris
Bacterial Gill Disease
Bacterial Endocarditis (*Lactobacillus*)
Peduncle Disease/Fin Rot

PARASITIC

Nodular Gill Disease
White Spot (Ichthyophthirius spp)
Costia
Tetrahymena (Dermal necrosis)
Lernea (Dermal necrosis)
Proliferative Kidney Disease
Corneal Diastomoniasis
Encephalic Diastomoniasis
Restrictive Pericardial Diastomoniasis

TOXIC

Nitrite toxicity

NUTRITIONAL

Starve out
Lipoid Liver Disease
Cataracts

ENVIRONMENTAL/MANAGEMENT

Hypoxia
Swimbladder Distress Syndrome (crowding)
Broken Spine (lightning/electrocution)
Gas Bubble Disease

IDIOPATHIC (unknown)

Gill Hyperplasia
Focal Hepatic Necrosis
Dorsal Aortic Thrombosis
Nephrocalcinosis

Fish used for experiments should be healthy. Although there are no set rules for the selection of healthy fish, a few guidelines are suggested.

(1) Where possible fish should be obtained from a certified source, although this is no guarantee that the fish are free of disease, it does indicate that a cursory health examination of the fish has been completed. (2) Growth appearance and behaviour of the fish should be normal. (3) Fish should be obtained from hatcheries on a spring water source, where possible, to decrease the chances of the fish having metazoan parasites. (4) Fish should be from hatcheries with good water quality and fish should have been fed a good diet. (5) An attempt should be made to obtain a diagnosis of any unexplained mortalities. (6) Where possible, a program of fish health surveillance should be used in fish stocks intended for experimental use.

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TOXICITY TESTING PROTOCOLS AND
SEPARATING THE MYTHOLOGY FROM THE MAJESTY

Gordon R. Craig

Abstract

Most provinces have either developed their own or adopted the federal toxicity test protocols for the regulation of industrial effluent discharges. They have been developed by different scientific groups over the last 15 years and, to a certain extent, reflect the level of understanding of the time.

Specific components among testing protocols will be compared and the individual limits for effluent transportation time, aeration, fish size, fish loading and concentration selection will be reviewed.

Emerging issues of pH control, dilution water effects, test reproducibility and biological variability will be addressed with respect to toxicity testing. The elements of good quality assurance programs will be outlined for better test characterization.

Finally, an objective review of the purposes of toxicity testing and inevitable effects of applying test results to environmental management programs will be discussed. Future needs in new regulatory biological test protocols will be identified.

Gordon R. Craig

RESUME

La plupart des provinces ont soit, développé leurs propre protocoles de test sur la toxicité, ou bien, ont adopté ceux du fédéral concernant la réglementation sur le déversement des effluents industriels. Ils ont été développés par différents groupes scientifiques durant ces 15 dernières années et, jusqu'à un certain point, ils reflètent l'évolution du niveau de compréhension dans le temps.

Des composants spécifiques parmi les protocoles des tests vont être comparés et les limitations individuelles des effluents, quant au temps de transportation, à l'aération, à la grosseur et quantité des poissons de même que quant à la sélection des concentrations, vont être révisées.

L'émergence des questions sur le contrôle du pH, les effets de dilution de l'eau, les tests sur la capacité de reproduction et les variantes biologiques vont être dirigés en considération des tests de toxicité. Les éléments assurant un programme de bonne qualité vont être décrits pour une meilleure caractérisation des tests.

Finalement, une révision objective de l'utilité du test de toxicité et les effets inévitables concernant l'application de ces résultats aux programmes de gestion de l'environnement vont être discutés. Les besoins futurs pour de nouveaux protocoles régularisant les tests biologiques vont être identifiés.

Activity, ventilation and physiological responses
of juvenile rainbow trout, *Salmo gairdneri*, to acid
and aluminum - prediction of field responses from laboratory data.
Christine Neville, Toxicity Unit, Ontario Ministry of the Environment

Juvenile rainbow trout were exposed to 75 ug/L inorganic aluminum at pH values from 6.5 to 4.0 with major ion concentrations similar to those found in the Muskoka-Haliburton district of Ontario. The severity of initial (1-2 hour) activity and ventilation responses predicted the severity of physiological responses after 6-11 days exposure, and indicated the possibility of detection and avoidance of similar conditions in the field. The initial response and physiological data can be used in conjunction with detailed knowledge of water chemistry fluctuations and the presence of refuge areas to predict the possibility of survival by tolerance, avoidance, or acclimation during similar toxic situations in the field. The sensitivity of other fish species can be estimated by comparison of the initial responses to those of rainbow trout during a few hours exposure to the same experimental conditions.

7555g

Abstract

Christine Neville

On a exposé les truites arc-en-ciel juvéniles à 75 ug/L d'aluminium inorganique dans des valeurs de pH de 6.5 à 4.0 avec de grandes concentrations d'ions semblables à ceux qu'on trouve dans la région de Muskoka-Haliburton de l'Ontario. L'intensité de la première activité et les réactions de ventilation des truites pendant les premières heures d'exposition (1 à 2 h) ont prédit l'intensité des réactions physiologiques après 6 à 11 jours d'exposition. Les premières réactions ont également indiqué qu'il existe la possibilité de déceler et éviter les mêmes conditions sur le terrain. On peut utiliser les premières réactions et les données physiologiques conjointement avec une bonne connaissance des variations de la chimie d'eau et l'existence des refuges pour prédire si les truites pourraient survivre en tolérant, en évitant ou en acclimatisant quand ils sont exposées aux mêmes conditions toxiques sur le terrain. En recréant les mêmes conditions expérimentales, on peut estimer la susceptibilité d'autres espèces de poissons en comparant les premières réactions après quelques heures d'exposition à celles des truites arc-en-ciel.

CANADIAN WATER QUALITY GUIDELINES REQUESTED BY THE CANADIAN COUNCIL OF
RESOURCE AND ENVIRONMENT MINISTERS

THE POSTER DESCRIBES THE ORIGINS OF THE CANADIAN WATER QUALITY GUIDELINES, THE PROCESS OF THEIR PRODUCTION, THEIR USE, AND THE PROPOSALS FOR RESEARCH TO PROVIDE ANSWERS TO FILL THE GAPS IN KNOWLEDGE THAT PREVENT. AT PRESENT, THE RECOMMENDATION OF GUIDELINES FOR MANY OF THE CHEMICALS WHICH AFFECT THE USE OF CANADA'S LAKES AND RIVERS.

A TEAM OF SCIENTISTS IS WRITING THE GUIDELINES USING MATERIAL, RELEVANT TO THE CANADIAN SITUATION, WHICH CAN BE FOUND IN REVIEWS ALREADY PUBLISHED. ONLY 12 MONTHS ARE AVAILABLE TO COVER 5 USES OF WATER: AQUATIC LIFE AND WILDLIFE; WATER SUPPLIES FOR DRINKING WATER; AGRICULTURE; RECREATION; AND INDUSTRY, FOR APPROXIMATELY 100 CHEMICAL, PHYSICAL AND BIOLOGICAL PARAMETERS WHICH COULD AFFECT THESE USES.

THE CCREM PUBLICATION WILL PROVIDE INFORMATION FOR LOCAL AGENCIES WHOSE RESPONSIBILITIES ARE TO SAFEGUARD THE QUALITY OF LAKES AND RIVERS IN THEIR JURISDICTIONS FOR WHATEVER-WATER USE IS REQUIRED. THE GUIDELINES WILL ENABLE SITE-SPECIFIC WATER QUALITY OBJECTIVES TO BE FORMULATED WITH AS MUCH ACCURACY AS POSSIBLE.

THERE ARE MANY GAPS IN THE NECESSARY KNOWLEDGE TO WRITE GUIDELINES AND TO USE THEM CORRECTLY, AND IT IS HOPED THAT THIS EXERCISE WILL ENABLE THE NECESSARY RESEARCH TO BE HIGHLIGHTED AND RECOMMENDED TO BOTH FUNDING AND RESEARCH AGENCIES IN THE PUBLIC AND PRIVATE SECTORS.

Margaret C. Taylor and Ronald C. Pierce.
Water Quality Branch,
Environment Canada.

Poster Abstract.

Margaret C. Taylor and Ronald C. Pierce

RESUME

L'affiche décrit les origines des lignes directrices sur la qualité de l'eau au Canada, le processus de leurs production, leurs usage, et les propositions de recherches visant à procurer les réponses qui serviront à combler les lacunes cognitives qui empêchent présentement la recommandation de lignes directrices pour plusieurs produits chimiques qui affectent l'usage des lacs et rivières du Canada.

Un groupe de scientifiques écrit présentement les lignes directrices utilisant le matériel, applicable à la situation canadienne, qui peut être trouvé dans les rapports déjà publiés. Douze mois seulement sont disponible pour couvrir ces 5 usages de l'eau: la vie aquatique et sauvage; les réserves d'eau potable; l'agriculture; la récréation; et l'industrie, pour approximativement 100 paramètres chimiques, physiques et biologiques qui peuvent affecter ces usages.

Les publications du CCMRE vont fournir de l'information pour les agences locales dont les responsabilités sont de sauvegarder la qualité des lacs et rivières sous leur juridiction peu importe l'usage requis de l'eau. Les lignes directrices vont permettre de formuler les objectifs sur la qualité de l'eau selon la spécificité du site avec la plus grande précision possible.

Il y a plusieurs lacunes quant à la connaissance nécessaire pour la rédaction de lignes directrices ainsi que pour leurs correctes utilisations, et il est espéré que cet exercice va permettre aux recherches nécessaires d'être soulignées et recommandées aux organismes de subventions et de recherches des secteurs public et privé.

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Endosulfan Toxicity and Residues in Tilapia nilotica and Carp

Little is known about the sensitivity of Tilapia species to pesticides and quantitative information is needed on their residues in the tissues of different fish. This information is needed to determine the cause of sudden mass mortalities or to prove suspected fish poisoning with the use of toxic agricultural chemicals.

The 72 hr and 96 hr lethal concentration (LC50) of endosulfan was determined for T. nilotica. Residues in different tissues were measured. Endosulfan residues in carp were measured after exposure to short-term lethal and long-term sublethal concentrations. 72 hr LC50 for Tilapia ranges between 2.61-3.06 ppb (95% CI 2.45-3.30), 96 hr LC50 between 2.05-2.79 ppb (95% CI 1.95-2.93). The gills contained between 542-2,151 ppb endosulfan which is about 5 to 10 times as much as the muscle which contained between 49-228 ppb. In carp, after short-term (up to 6 hrs) lethal exposures, body and head contained an average of 630 ppb endosulfan (216-1,330 ppb), gills 1,106 ppb (401-2,740 ppb), liver and bile 1,056 ppb (216-2,055 ppb) and the intestines 1,045 ppb (554-2,355 ppb). After long-term (55 days) exposure to sublethal concentration, carp muscle contained an average of 24 ppb endosulfan (11.7-37.5 ppb), gills 79 ppb (54.3-91.5 ppb), liver and bile 339 ppb (76.7-880 ppb), intestines 64 ppb (21.5-246 ppb). No development of a resistance against endosulfan could be observed in carp.

Abraham M. Herzberg

RESUME

On en sait très peu sur la sensibilité des espèces *Tilapia* face aux pesticides et, une information quantitative est nécessaire sur leurs résidus dans les tissus de différents poissons. Cette information est nécessaire pour déterminer la cause des mortalités soudaines de masse ou pour confirmer les doutes concernant le braconnage aquatique avec l'usage des produits chimiques toxiques utilisés en agriculture.

La concentration mortelle (LC50) d' "endosulfan" pour une période de 72 hr. et 96 hr. a été déterminée pour *T. nilotica*. Les résidus contenus dans les différents tissus ont été mesurés. Les résidus d' "endosulfan" chez la carpe ont été mesurés après une courte période d'exposition mortelle et après une longue période d'exposition au-dessous du niveau mortel de concentration. 72 hr LC 50 pour *Tilapia* varie entre 2.61 - 3.06 ppb (95% CI 2.45 - 3.30), 96 hr LC50 entre 2.05 - 2.79 ppb (95% CI 1.95 - 2.93). Les branchies contenaient entre 542 - 2,151 ppb d' "endosulfan", ce qui est environ 5 à 10 fois plus que les muscles, qui eux, contenaient entre 49 - 228 ppb. Chez la carpe, après une courte période (jusqu'à 6 hr.) d'exposition mortelle le corps et la tête contenaient une moyenne de 630 ppb d' "endosulfan" (216-1, 330 ppb), les branchies 1,106 ppb (401-2, 740 ppb), le foie et la bile 1,056 ppb (216-2, 055 ppb) et les intestins 1,045 ppb (554-2, 355 ppb). Après une longue période (55 jours) d'exposition au-dessous du niveau mortel de concentration les muscles de la carpe contenaient en moyenne 24 ppb d' "endosulfan" (11.7 - 37.5 ppb), les branchies 79 ppb (54.3 - 91.5 ppb), le foie et la bile 339 ppb (76.7 - 880 ppb), les intestins 64 ppb (21.5 - 246 ppb). Aucun développement de résistance contre l' "endosulfan" a été observé chez la carpe.

USING ECOTOXICOLOGICAL TESTING FOR
DREDGE SPOIL HAZARD ASSESSMENT
UNDER THE OCEAN DUMPING CONTROL ACT (ODCA).

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K1A 1C8

Chevrier, A. and J. Karau, 1985. Using ecotoxicological testing for dredge spoil hazard assessment under the Ocean Dumping Control Act (ODCA).

ABSTRACT:

Considerable research work has been conducted to date on tests for assessing ecotoxicological effects of marine dredge spoils.

The Ocean Dumping Program of EPS funded some of this work and is now involved in the development of a framework within which selected tests can be used to conduct dredge spoil hazard assessments.

We are primarily concerned with contaminant effects at dumpsites. In order to answer this environmental concern we are undertaking a review of ecotoxicological tests and criteria that could be used for decision-making under the ODCA.

R  SUM  :

Beaucoup d'efforts ont   t   d  di  s jusqu'   maintenant,    la recherche reli  e    des tests permettant d'  valuer les effets   cotoxicologiques des s  diments dragu  s marins.

Le programme d'immersion du SPE a investi des fonds dans ces recherches et se penche maintenant sur le d  veloppement d'un cadre    l'int  rieur duquel des bio-tests choisis peuvent   tre utilis  s pour l'  valuation des mati  res dragu  es.

Le programme est surtout int  ress   par les effets des contaminants aux nouveaux lieux d'immersion. Afin de r  pondre    cette question environnementale, le programme s'est engag   dans une revue des tests et crit  res   cotoxicologiques pouvant   tre utilis  s dans le cadre de la loi d'immersion.

INTRODUCTION

EPS is responsible for administering the ODCA which pledges that Canada prevents marine pollution by dumping and controls of all sources of marine pollution. Ocean disposal of dredge spoil is the major dumping activity permitted under the Act (approximately 13,000,000 m³ or 20,000,000 tonnes annually). One of the major environmental concerns is that toxic substances contained in the sediments may pose an environmental threat when dredging or dumping remobilizes them. On the average, the permit applications show that less than 10% of the dredge spoil intended for ocean disposal is considered to be highly contaminated.

Consequently, the potential adverse (acute and/or chronic) effects resulting from the ocean disposal of dredge material containing toxic substances must be evaluated through the regulatory review process prior to approval of ODCA permits.

EPS has, therefore, identified the need to improve and expand the role of toxicological results in addressing the regulatory questions:

- to what extent will dumping at sea cause acute adverse effects to representative marine organisms at the dump site, and can these be minimized?
- to what extent will dumping at sea cause significant irreversible chronic effects to representative marine organisms at the dump site, and can these be minimized?

In order to select tests which would address the above questions, EPS has initiated a feasibility assessment of sediment toxicity tests suitable for ODCA dredging application reviews.

The object of this paper is to report on the feasibility assessment study design and objectives and explain what factors were taken into account. The study outline was prepared in the context of a scientific and regulatory environment which will be described first.

REGULATORY ENVIRONMENT FOR DREDGING

The 1982 study "Review of Disposal Alternatives for Dredged Material" recommended that the following should be the primary goals reflected in any reform of the dredging regulatory process:

- ° Ensure that the scientific testing methods utilized to estimate the environmental impact of sediment disposal keep up with technological advances and are appropriate to the sediments being disposed.
- ° Apply across Canada, a standardized review approach for evaluating proposed dredging projects and disposal alternatives, recognizing that within such standardization, there will be variation within specific guidelines to allow for differences between ocean, Great Lakes and other fresh water projects."

The attached flow chart of dredging project evaluation is from the report of the IJC Dredging Sub-Committee, January 1982, and outlines the sequences of assessments that are generally used for regulatory purposes in Canada and abroad. In addition, the London Dumping Convention (LDC), which lead to the enactment of the ODCA in Canada, issued guidelines (LDC 8/10, Annex 2) on test procedures to be used by contracting parties to evaluate whether material dumped at sea will be "rapidly rendered harmless". The guidelines call for:

- a) acute toxicity tests on plankton, crustaceans or molluscs, and fish;
- b) chronic toxicity tests capable of evaluating long-term sub-lethal effects, such as bioassays covering an entire life cycle;
- c) tests to determine the potential for bioaccumulation and, if appropriate, the potential of elimination; and
- d) tests for determining the persistence of substances.

In line with the LDC guidelines and the generally applied regulatory process for dredged material, the ODCA requires that all dredge material be evaluated prior to dumping at sea to ensure the material will be "rapidly rendered harmless". Currently, a permit for dredge disposal may be issued if the concentration of any substance in the dredge material is not in excess of the ODCA regulated limits or the local baseline concentrations at the dump site. If the dredge material fails these chemical screening tests, a permit may still be issued if biological tests show that dumping the dredge material will not cause acute or irreversible chronic effects in sensitive marine organisms typical of the marine ecosystem at the dump site. At present, the ODCA does not prescribe any biological tests to evaluate acute or chronic biological effects.

SCIENTIFIC ENVIRONMENT

Some ODCA research funds have been dedicated over the years to the development of tests for assessing ecotoxicological effects of marine dredge spoils hazards. A roll-up of research results generated between 1975-1982 lead to the following conclusions:

- 1) Concerns exist on a site specific basis, that toxics contained in contaminated sediments may be taken up by biota and potentially endanger marine life or human health.
- 2) The remobilization and bio-availability of toxics from dredged sediments is only partially understood. The main concern lies with contaminants remobilized in overlying and interstitial water. However, the toxicological consequences of the release are not well documented.

- 4) The data on bio-availability suggests that uptake is mainly from the aqueous phase. Sediments appear to bind substances to particulates or produce insoluble compounds which serve to reduce the availability of contaminants. No research results are available on acute or chronic toxic effects of contaminated sediments.

Taking into account ODCA research results and other sources, EPS developed the following premises to the proposed ODCA study on use of ecotoxicological testing:

- 1) Research on the fate and effects of dredge spoil disposal in the marine environment indicates that many of the toxic substances in dredge spoil are available to and potentially toxic to benthic and epibenthic organisms.
- 2) Many of these organisms live in and feed on dredge spoil and, consequently, are considered to be the most appropriate organisms in solid and suspended particulate phase bioassays.
- 3) Since acute lethality from exposure to contaminated sediment is seldom observed, attention must also be directed towards the assessment of chronic and sublethal effects.
- 4) Regulatory tests need to be sensitive and precise. The responses measured in the regulatory tests should be sensitive to specific classes of chemicals (e.g., organohalogens), and should not be sensitive to expected variations in other variables (i.e. low S/N ratio).
- 5) The dredge material bioassay is expected to provide a quantitative prediction of the toxic effects of the dredge material, essential for a hazard assessment of the material at the dump site.
- 6) Field verification of hazard assessment predictions is also highly recommended.

STUDY RELATED TO ECOTOXICOLOGICAL TESTING UNDER THE ODCA

Using the information presented above, EPS designed a study to conduct a feasibility assessment of sediment toxicity tests suitable for ODCA dredging application reviews with the following objectives:

- 1) Provide a feasibility assessment of marine sediment toxicity tests which address the ODCA dredging application review question will dumping dredge spoil at sea cause acute or irreversable chronic effects in sensitive marine organisms typical of the marine ecosystem at the dump site?

- 2) Recommend toxicity tests that can be used to help make ODCA decisions regarding the environmental acceptability of dumping contaminated dredge spoil at sea.

The study will cover the following area:

- 1) Review and critically evaluate the biological testing procedures for assessing the toxic effects of dredge spoil disposal in the marine environment. The factors and criteria to be included in the review are identified in Table II (attached).
- 2) Evaluate the test procedures currently used or recommended by contracting parties to the London Dumping Convention (LDC) as well as those funded by the ODCA research program.
- 3) The primary scientific literature should be consulted in the review.
- 4) Recommend tests, according to the criteria in Table II, which should be considered alone or in combination for:
 - immediate laboratory use as a quantitative prediction of the toxic effects of dredge spoil disposal at sea,
 - immediate field use to verify hazard assessment predictions, and
 - further development or trial use.
- 5) Provide examples of the results and the pass/fail criteria used or which could be used for the recommended toxicity tests.

CONCLUSION

Results of the study will be available in April 1986 and help ODCA program managers in refining the hazard assessment of dredge material by defining in the form guidelines, which ecotoxicological tests and pass/fail criteria should be employed.

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TABLE 1: FLOW CHART OF DREDGING PROJECT EVALUATION

(From: Dredging Sub-Committee, Water Quality Programs Committee of the Great Lakes Water Quality Board (1982) Guidelines and Register for Evaluation of Great Lakes Dredging Projects).

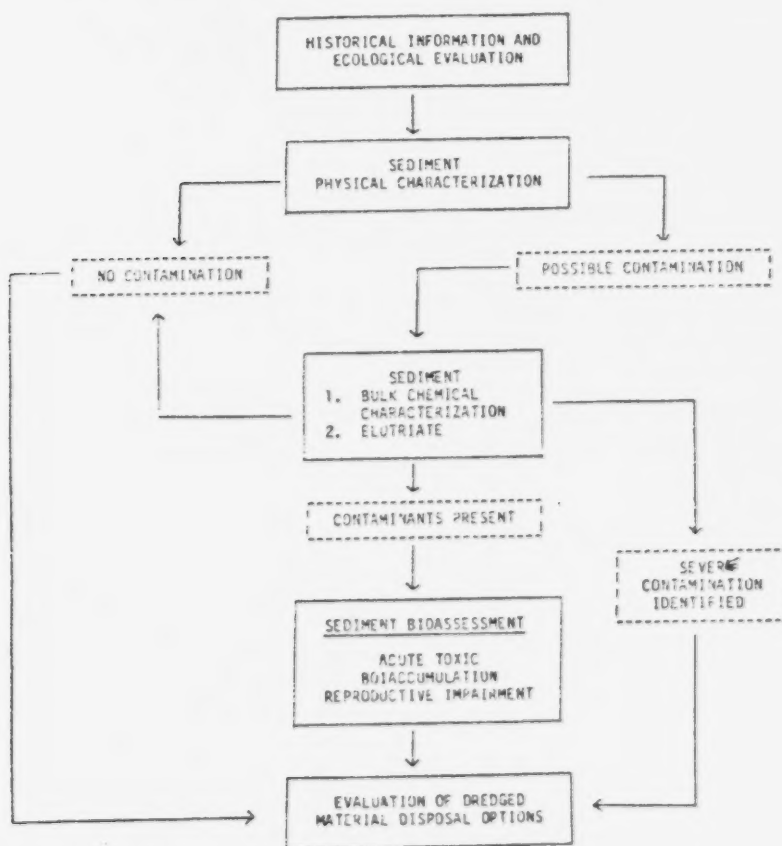


TABLE II: FEASIBILITY ASSESSMENT OF SEDIMENT TOXICITY TESTS
SUITABLE FOR ODCA DREDGING APPLICATION REVIEWS

FEATURES	CRITERIA
Test Organism(s)	- Benthic and epibenthic marine organisms typical of ocean dump sites - Can be reared in the laboratory or easily collected and maintained - Biological and toxicological information base exists for the organism(s)
Responses 1) Test Conditions 2) Toxicological Criteria 3) Sensitivity	- Solid phase - Suspended particulate phase - Lethal and sub-lethal responses for single and multiple species tests - The effect can easily be detected above the natural variability
Practical 1) Rapidity Evaluation 2) Cost/Test 3) Training Equipment, Expertise	- Measurable effects within days or up to 2 weeks - Moderate cost - Capable of being carried out by government and commercial labs
Application	- The effect can be used to predict adverse acute and chronic effects at the dump site - The effect has been or could be verified in a field monitoring program

Abstract

Toxicity/Time Relationships for Fathead Minnows (Pimephales promelas)
Exposed to PesticidesJarvinen, A. W. and D. K. Tanner
ERL-Duluth, Minnesota

A continuing pesticide research project conducted at the Environmental Research Laboratory-Duluth, MN (ERL-D) involves the determination of toxicity/time relationships for fathead minnows and four pesticides; chlorpyrifos, an organophosphate; fenvalerate, a synthetic pyrethroid; endrin, a chlorinated hydrocarbon; and triphenyltin hydroxide, an organotin. Exposure regimes observed in the field are different from those normally used in laboratory bioassays where aquatic organisms are exposed to a constant pesticide concentration in the water over the studies pre-set exposure duration. To validate laboratory-derived toxicity data for comparison to field data, the development of toxicity/time relationships is necessary. Completed bioassay results for continuously exposed larval fathead minnows with chlorpyrifos demonstrate:

- 96 hour LC50 of 120 $\mu\text{g/l}$
- 24 + 8 hour LC50 of 400 $\mu\text{g/l}$
- 4 hour LC50 of 782 $\mu\text{g/l}$
- 2 hour or less requires a concentration greater than water solubility ($>1000 \mu\text{g/l}$)

A continuous exposure concentration that caused 50 percent deformities in the test fish was 55 $\mu\text{g/l}$. A similar response occurred at a 1-hour exposure to 761 $\mu\text{g/l}$, 2 hours at 365 $\mu\text{g/l}$, 4 hours at 216 $\mu\text{g/l}$, 8 hours at 184 $\mu\text{g/l}$ and 24 hours at 30 $\mu\text{g/l}$. Results of 30-day studies show that a statistically significant increase in deformities and decrease in growth occur if the fish are exposed for 5 hours to a concentration similar to a continuous exposure 96-hour LC50 value (120 $\mu\text{g/l}$). A 1-hour exposure at twice the LC50 value caused similar results.

La relation toxicité/temps pour les vairons
à "grosse tête" (Pimephales promelas) exposés aux pesticides.

Un projet de recherche continu sur les pesticides effectué au Laboratoire de Recherche Environnementale de Duluth, MN (LRE-D) implique la détermination du rapport toxicité et temps pour les vairons à "grosse tête" et quatre pesticides; le "chlorpyrifos" un phosphate organique; le "penvalérate", un "pyréthroïde" synthétique; l'endrin, un hydrocarbure chloruré; et l'hydroxide "triphenétin", un "organotin". Les régimes d'exposition observés sur le terrain sont différents de ceux normalement utilisés lors des tests biologiques de laboratoires où les organismes aquatiques sont exposés à une concentration constante de pesticides contenus dans l'eau comparativement aux études de durée d'exposition pré-établie. Pour rendre valide les données sur la toxicité issues des laboratoires aux fins de comparaisons avec celles sur le terrain, le développement de la relation toxicité/temps est nécessaire. Les résultats des tests biologiques complétés pour les larves des vairons à "grosse tête" exposés de façon continuë au "chlorpyrifos" démontrent:

- 96 heures LC 50 de 120 µg/l
- 24 + 8 heures LC 50 de 400 µg/l
- 4 heures LC50 de 750 µg/l
- 2 heures ou moins requiert une concentration plus grande que la solubilité de l'eau (>1000 µg/l)

Une exposition continue à une concentration causant 50% de difformité chez les poissons utilisés pour l'expérience était de 55 µg/l.

Une réponse similaire survient lors; d'une exposition d'une heure à 761 µg/l, de 2 heures à 366 µg/l, de 4 heures à 216 µg/l, de 8 heures à 184 µg/l et de 24 heures à 30 µg/l. Les résultats d'études effectuées sur une période de 30 jours démontrent statistiquement un accroissement considérable de la difformité et une diminution de la croissance survient si les poissons sont exposés durant 5 heures à une concentration similaire à une exposition continue de 96 heures à une valeur de LC50 (120 µg/l). Une exposition d'une heure au double de la valeur de LC50 cause des effets similaires.

AN IN SITU TOXICITY STUDY OF A PULP AND PAPER MILL
EFFLUENT AFFECTING JACKFISH BAY, LAKE SUPERIOR

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An in situ toxicity study of a pulp and paper mill effluent affecting
Jackfish Bay, Lake Superior. Can. Tech. Rep. Fish. Aquat. Sci.

An exposure of rainbow trout (Salmo gairdneri) was undertaken in
Jackfish Bay to determine the zone of impact of a pulp mill waste
in July, 1983. The mill is a bleached kraft operation which
discharges up to 138,000 cu m of effluent per day into Moberly Bay
(the western arm of Jackfish Bay) via Blackbird Creek.

Within two days, complete fish mortality had occurred at all cage
sites in Moberly Bay and as far as 1.5 km down the western shore
to Cody Island. Dissolved oxygen levels in the zone of impact
at 48 h, were below the MOE objective for cold water fish of 5 mg/L
at 20 C. In addition, toxic components such as resin acids and
chlorophenols were measured in the effluent plume. No further
significant mortality occurred after the 48 h observation.

Water samples from Jackfish Bay were found to contain detectable
levels of 2,4,6 trichlorophenol and pentachlorophenol in the ppt
range. The whole body analyses of fish which survived the 96 h
exposure at a site 1.5 km from the Blackbird Creek discharge,
demonstrated the bioconcentration of 2,4,6 trichlorophenol (in the
ppb range).

Concurrent with the field study, 96-h bioassays to measure the
acute lethality of the mill influent, effluent and Blackbird Creek
were performed at the MOE Thunder Bay laboratory. Based on the
conductivity at the field sites relative to the Blackbird Creek
discharge, the estimated effluent concentration at 4 of 6 fish
exposure sites with complete mortality ranged from 40-46% at 48 h.
Those values are consistent with the laboratory result of complete
mortality in the 45% test solution and an LC50 of 37% for that day's
Blackbird Creek sample.

In conclusion,

1. The zone of impact consisted of Moberly Bay and the western shore
of Jackfish Bay, 1.5 km from the Blackbird Creek discharge.
2. The fish lethality was due to a combination of effluent consti-
tuents and related D.O. levels.
3. The laboratory and field bioassay results were comparable.

4. Chlorophenol uptake by fish was a localized effect which occurred within 96 h.

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FLOOD, K., D. HOLLINGER, M. THOMSON, W. WAGER & BANAS. 1985.
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RESUME

En juillet 1983, nous avons étudié l'exposition des truites arc-en-ciel (*Salmo gairdneri*) dans la baie Jackfish pour déterminer la largeur de la zone affectée par les déchets d'un moulin à papier. Le moulin est une opération de kraft blanchie qui rejette jusqu'à 138.000 cu m d'effluent chaque jour dans la baie Moberly (le bras de la baie Jackfish de l'ouest) en passant le ruisseau Blackbird.

En moins de deux jours, nous avons trouvé un taux de mortalité de 100% sur tous les sites étudiés dans la baie Moberly, jusqu'à l'île Cody à 1.5 km le long de la côte ouest. Après 48 h, la quantité d'oxygène dissoute dans la zone affectée n'a pas atteint le niveau acceptable de 5mg/L à 20 °C du ministère de l'environnement ontarien. De plus, nous avons décelé des composants toxiques comme par exemple, des résines d'acides et des chlorophénols dans la plume d'effluent. Au bout de 48 h d'observation, nous n'avons pas trouvé de changement significatif du taux de mortalité.

Des niveaux discernables de 2,4,6 trichlorophénol et pentachlorophénol ont été trouvés dans la gamme de p.p.t. dans les échantillons d'eau de la baie Jackfish. L'analyse complète des poissons qui ont survécu après une exposition de 96 h à un site à 1.5 km de la décharge du ruisseau Blackbird a démontré une bioconcentration de 2,4,6 trichlorophénol (dans la gamme de p.p.b.).

Pendant que nous faisons cette étude, le laboratoire du ministère de l'environnement de l'Ontario de Thunder Bay a fait des essais biologiques d'une durée de 96 h pour mesurer la létalité aigue du ruisseau Blackbird et des influents et effluents du moulin. Basée sur la conductivité des sites étudiés, la concentration d'effluents estimée dans 4 sites sur 6 avec un taux de mortalité de 100% allait de 40% à 46% après 48 h. Les résultats sont compatibles avec les résultats de l'étude en laboratoire qui a démontré un taux de mortalité de 100% dans une solution d'analyse composée de 45% d'effluents et d'un LC50 de 37% de l'échantillon du ruisseau Blackbird ce jour-là.

En conclusion,

1. La baie Moberly et la côte de l'ouest de la baie Jackfish à 1.5 km de la décharge du ruisseau Blackbird était la zone affectée.
2. La létalité des poissons a été provoquée par une combinaison des constituants d'effluents et du niveau d'oxygène dissoute.
3. Les essais biologiques en laboratoire et sur le terrain était comparables.
4. L'absorption des chlorophénols par les poissons était un effet localisé qui s'est produit en moins de 96 h.

